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Amphibian population declines in montane southern Mexico: resurveys of historical localities

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Abstract

Declines of amphibian populations have been well documented in the US, Canada, and Central America, but little is known regarding the status of amphibian populations in Mexico. In 2000, we surveyed 43 transects from 3 upland regions in Guerrero and Oaxaca, Mexico. We found 161 adult amphibians belonging to 39 species, representing only 19–48% of the anuran fauna known from these regions. We found one dead (*Eleutherodactylus saltator*) and one dying frog (*Ptychohyla erythromma*) from two different streams near Chilpancingo, Guerrero. Both frogs were infected with *Batrachochytridium dendrobatidis*, a pathogenic fungus involved in other declines of amphibian populations. We collected 368 tadpoles; 60 (19%) tadpoles from 9 different streams among the three regions were missing mouthparts, which is indicative of infection by chytrid fungus. We report additional data from the state of Chiapas, Mexico, that document declines, disease, and apparent extirpations from that region as well. Overall, we report 31 populations (representing 24 species) that appear to have been extirpated, including populations of as many as 11 endemic species that have been missing for 16–40 years and may be extinct.

Keywords: Amphibians; Declines; Mexico; Montane; Population

1. Introduction

Populations of amphibians across the globe have been declining for many years (Alford and Richards, 1999; Houlahan et al., 2000). Many declines of amphibians in Latin America have occurred in upland areas (⩾600 m elevation) since the early 1980s (Young et al., 2001); some of these have affected half the species in any given region (Lips et al., 2003b). This phenomenon is particularly evident in species associated with streams (Pounds et al., 1997; Lips, 1998, 1999). Given the high endemism and restricted ranges of many Latin American species (Campbell, 1999; Duellman, 1999; McCranie and Wilson, 2002; Savage, 2002), some of these species may now be extinct. Although habitat loss and alteration have resulted in the direct loss of some amphibian populations, they cannot explain the decline of populations in relatively pristine habitats and protected reserves.

Hypothesized causes of amphibian declines in Latin America are varied, but include habitat loss, climate change (patterns of temperature, rainfall, and UV-B radiation), disease (chytrid fungi, ranavirus), exotic species (predators, competitors, pathogens), chemical contamination (toxins, endocrine disruption), and potential interactions among several factors (reviewed in Young et al., 2001). Some amphibian declines in Latin America (e.g., Costa Rica: Lips et al., 2003a; Panama: Berger et al., 1998; Lips, 1999; Ecuador: Ron et al.,

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2003) have been attributed to infection by *Batrachochytrium dendrobatidis*, an amphibian-specific aquatic, chytrid fungus (Longcore et al., 1999).

In Central America, anuran declines have been documented from upland cloud forests in Guatemala (Campbell, 1998), Honduras (McCranie and Wilson, 2002), Costa Rica (Pounds et al., 1997; Lips, 1998; Lips et al., 2003b), and Panama (Lips, 1999). Amphibian declines have also been reported from several areas in the western United States (e.g., Knapp and Matthews, 2000; Pilliod and Peterson, 2001; Jaeger et al., 2001; Bradley et al., 2002). Despite numerous documented anuran declines in North America and Central America, most reports from Mexico have been anecdotal (e.g., Scott, 1993; Santos-Barrera et al., 1994; J.A. Campbell, pers. comm.). An exception is the study by Hale and Jarchow (1988), who conducted a resurvey of historical localities of *Rana tarahumarae* in southern Arizona and northern Mexico. This frog had been regularly encountered throughout the region from its discovery in the 1930s until 1974 (Hale and Jarchow, 1988). By 1980, all Arizona populations had disappeared and, within a few years, two northern Mexico populations had declined (Hale and May, 1983). In Sonora, Mexico, between 1981 and 1986, Hale and Jarchow (Hale and Jarchow, 1988) reported finding dead and dying frogs from several areas and concluded that pollution from copper smelters might have contributed to declines, although toxicological analyses of frogs and water produced no evidence of chemical poisoning. The actual cause of the decline of *R. tarahumarae* is not known, but Bradley et al. (2002) documented chytrid infections in *Rana yavapaiensis* and *Rana chiricahuensis* as early as 1992 in southern Arizona and chytrid fungus also has

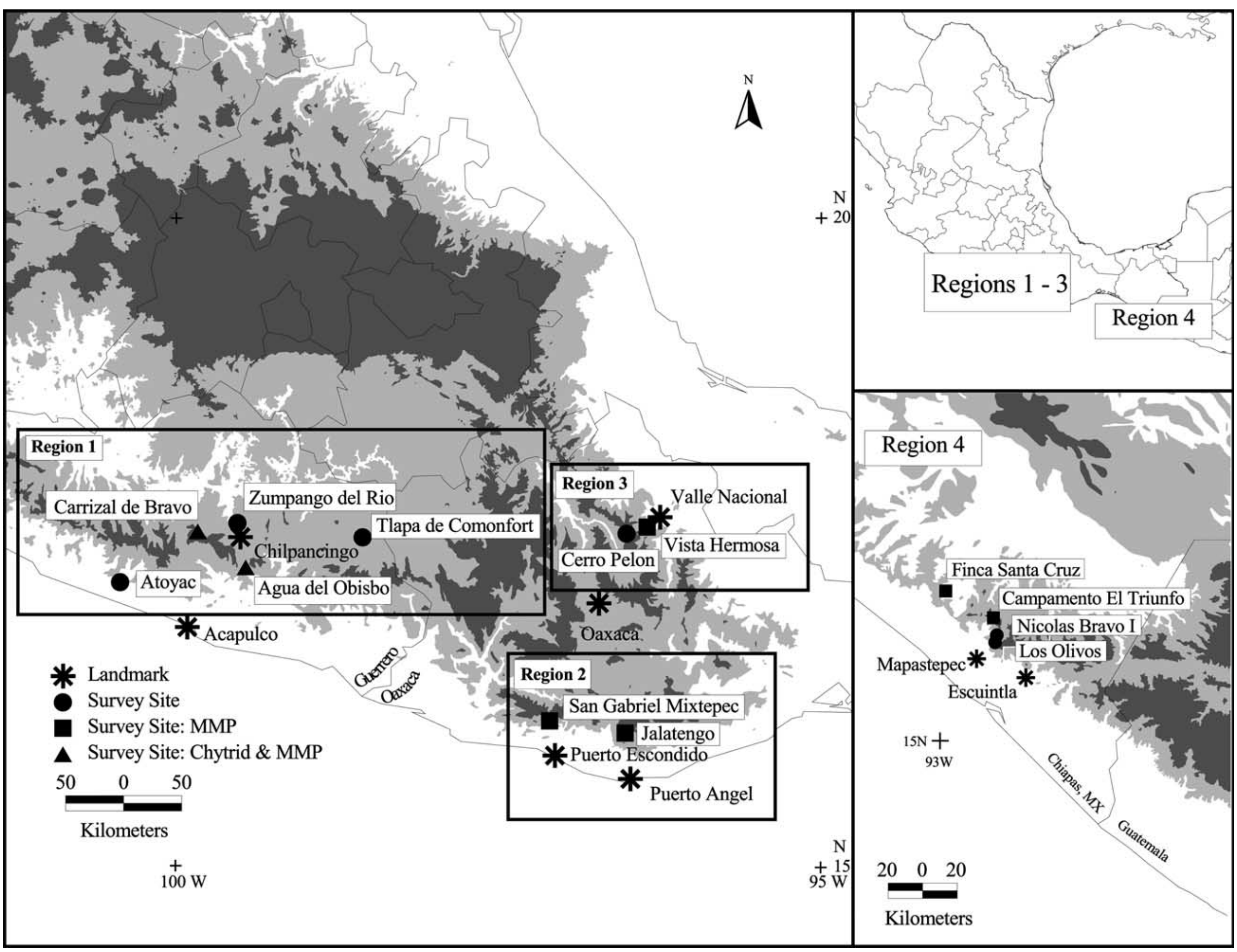

Fig. 1. Generalized map of southern Mexico, showing the study regions in the states of Guerrero, Oaxaca, and Chiapas, which were surveyed in this study. Survey sites represent more than one individual transect or site, and are labeled as the closest towns nearby (see text). We found indirect (tadpoles with missing mouthparts: MMP) and/or direct (from diagnostic exams of dead frogs: Chytrid) evidence of the presence of chytrid fungus in all three regions. Larger cities are given as landmarks, while survey sites are labeled according to evidence of presence/absence of the fungus. White areas indicate elevations between 0 and 900 m, pale gray indicates 900–2100 m, and dark gray indicates elevations above 2100 m. Regions 1–3 are shown on the left; Region 4, near the Chiapas, MX-Guatemala border, is shown in the lower left, where all study sites are located within the El Triunfo Biosphere Reserve. Upper right corner shows the general area in Mexico where regions are located; symbol legend and shaded relief contours are the same for Regions 1–4.

been found in specimens of *R. tarahumarae* collected as early as 1978 (S. Hale, C. Goldberg, pers. comm.).

Mexico has the greatest number of amphibians (318 species) and the greatest number of endemic anurans of any Central or North American country (Campbell, 1999), therefore there is an urgent need to evaluate the status of Mexican amphibian populations. Because Mexico is geographically intermediate to sites with documented declines in North America and Central America, we hypothesized that declines had already occurred in Mexico. We proposed to evaluate the diversity and abundance of stream amphibians in four regions (Fig. 1) that are topographically and ecologically similar to other sites of decline. We targeted upland sites with relatively unaltered original habitat, and specifically cloud forest stream frogs and their larvae as those most likely to show obvious declines. We revisited sites that had been extensively surveyed in previous years from which we established historical presence/absence of species in these areas. We quantified the species richness and abundance of anurans along streams at 1–5 localities in each of the four regions, and gathered material to determine the presence of the fungus, *B. dendrobatidis*. We did not collect information on the presence of exotic predators, UV radiation, iridiovirus, chemical contaminants, climate change, or any other factors potentially related to amphibian declines; as a result, we can only comment on the presence of chytrid fungus and large-scale habitat modification.

2. Methods

2.1. Study regions

2.1.1. Region 1: Pacific Slope Sierra Madre del Sur, Guerrero (600–2300 m elev.; Fig. 1)

This area is characterized by humid, semideciduous forest, cloud forest, or pine-oak-fir forest with cascading, perennial forest streams. The area from Atoyac north to Puerto del Gallo and Carrizal del Bravo has been surveyed by various collectors – especially K. Adler in the 1960s (Adler, 1965, 1996; Adler and Dennis, 1972), field crews from University of California Berkeley in the 1970s and 1980s (e.g., Papenfuss et al., 1983; Wake et al., 1992), and J.A. Campbell and associates from The University of Texas at Arlington (UTA) from the 1970s to the present. The area south of Chilpancingo, especially near the private hacienda of Agua del Obispo, has been repeatedly surveyed by Taylor (1930s and 1940s), Smith (1940s), Duellman (1950s and 1960s) and Dixon (Davis and Dixon, 1965). Based on these collections, and museum records, we generated a list of 36 anuran species known from this region (27% endemic species). Between 27 June and 3 July, 2000, we conducted 13 surveys in this region, 2 north of Atoyac, 3 near Carrizal del Bravo, 6 near Agua del Obispo, one north of Zumpango del Río, and 1 west of Tlapa de Comonfort.

2.1.2. Region 2: Pacific Slope Sierra de Miahuatlan, Oaxaca (700–1400 m elev.; Fig. 1)

This region is characterized by humid, semideciduous forest, cloud forest, or pine-oak-fir forest with cascading, perennial forest streams. Caldwell (1974a) intensively studied the habitat associations of hylid treefrogs in the area north of San Gabriel Mixtepec and near the Río Jalatengo between 1969 and 1970, providing historical estimates of species richness and abundance for numerous streams in these two areas. We generated a list of 31 anuran species from this region (26% endemic species). Between 5 and 10 July, 2000, we conducted 8 surveys centered around these two portions of Caldwell's transects: 5 surveys along the streams above San Gabriel Mixtepec and 3 surveys of streams near the Río Jalatengo.

2.1.3. Region 3: North Slope Sierra de Juárez, Oaxaca (1300–2400 m elev.; Fig. 1)

This area is characterized by rain forest, cloud forest, and pine-oak forest with cascading, perennial forest streams. Caldwell (1974a) intensively studied the habitat associations of hylid treefrogs in this region between 1969 and 1970, providing historical estimates of species richness and abundance. Additional, extensive collections have been made by W.E. Duellman and field crews from The University of Kansas (KU; 1960s) and by J.A. Campbell and associates from UTA (1970s and 1980s); other collections have been made in the 1990s by JRM and field crews from the Museo de Zoología, Universidad Nacional Autónoma de México (UNAM). We generated a list of 25 anuran species from this region (26% endemic species). Between 11 and 15 July, 2000, we conducted 17 surveys along numerous streams between Cerro Pelón and Vista Hermosa, which represented the third portion of Caldwell (1974a) transect.

2.1.4. Region 4: Pacific Slope Sierra Madre, Chiapas; Reserva de la Biosfera EL Triunfo. (600–2100 m elev.; Fig. 1)

This region is characterized as montane and premontane rainforest, and cloud forest, with numerous permanent streams. This is a protected region, but in some areas the forest is interspersed with plots of coffee. Between 1938 and 1940, Taylor and Smith (1945) collected in the low- to mid-elevations of this area (500–1500 m). Between 1993 and 1994, Luna (1997) studied the abundance of amphibians and reptiles of the higher elevations (1500–2100 m). Additional collections in this region include work by: N. Hartweg of the University of Michigan (UMMZ) in 1947; J.M. Legler of the Field Museum (FMNH) in 1940; Miguel Álvarez del Toro of Tuxtla Guittierez, Chiapas, in 1960; teams from the

Institute of Natural History and Ecology of Chiapas (IHN) in 1994; and Muñoz from Colegio de la Frontera Sur (ECOSUR) in 1997–2002. These collections have identified 29 species of frogs from this area (21% endemic species). We (AMA and associates) surveyed 13 transects in the reserve: 4 in the El Triunfo camping area (2050 m), 2 at Nicolas Bravo I (1200–1300 m), 3 at Finca Santa Cruz (1300 m), and 4 at Los Olivos (620–765 m).

2.2. Field methods and analyses

Regions 1–3 were visited by teams of 3–6 herpetologists (always including KRL, JRM, and DGM), sampling multiple localities within each region for 4–6 days during the rainy season between June and July, 2000 (see Study regions for details). In each region we conducted diurnal and nocturnal visual encounter surveys (Heyer et al., 1994) of multiple streams and trails where possible. We captured and identified (species, sex, approximate age) all individual amphibians and reptiles encountered, and collected voucher specimens of adults and tadpoles of all species for identification and diagnostic exams. When we encountered breeding choruses we estimated the number of calling males. We opportunistically collected tadpoles from all streams and pools by dipnetting representative microhabitats (e.g., riffles, pools, under rocks, in vegetation). All specimens collected were deposited at UTA and UNAM. We compared our survey results to historical records to determine if there was evidence of declines of anuran populations, which we defined as the absence of species previously common and whether there was any evidence of either habitat loss or chytrid fungus. Surveys in Region 4 (Chiapas) were conducted independently by teams of students and faculty from ECOSUR, led by AMA; field work was conducted during the rainy seasons of 1999 and 2001.

In Regions 2 and 3 we evaluated changes in amphibian capture rates from the same stream transects surveyed by Caldwell 25 years previously (Caldwell, 1974a). In Regions 1 and 4 we were only able to compare the number of species we encountered to historical collections. Caldwell (1974a) surveyed over 200 transects between September 1969 and August 1970, studying the ecology of 35 hylid species along a 3000 m elevational transect. She surveyed 1–45 transects per month, with the greatest intensity between March and July 1970 (16–45 transects/mo). Caldwell found as many as 7 species and 400 individuals in a given survey during her visits to these sites, but did not record any frog captures in approximately 20% of her transects. She may have seen or heard frogs during those surveys, but her field notes did not indicate so. Caldwell collected adults and/or larvae of most hylid species throughout the year, suggesting that they have prolonged breeding seasons and/or long larval periods (Caldwell, 1974a); based on this, we expected to find adults or larvae of most hylid species during our surveys in June and July.

We examined the condition of mouthparts on all preserved tadpoles with a variable power dissecting microscope. We coded each tadpole as to whether the jaw sheaths and tooth rows were complete or whether half or more of each of these structures were missing. We did not quantify mouthpart condition for any individuals at Gosner (1960) Stages 40–42, because of the likelihood that this was a natural loss seen during metamorphosis.

We calculated the simple linear regressions between elevation and average capture rate for all transects combined, and for the nocturnal stream transects alone. We used nonparametric Wilcoxon/Kruskal–Wallis rank sum tests to compare differences in capture rates between terrestrial and riparian habitats, and between the number of both captures and species found by Caldwell in 1970 and by us in 2000. All statistical analyses were conducted in the program JMP (SAS Institute, 2000).

3. Results

Between 19 June and 25 July, 2000, we conducted 43 surveys for anurans in Regions 1–3 (Guerrero and Oaxaca, Mexico); additional attempts were made to conduct surveys in the Sierra Madre Oriental, in the State of Veracruz. In Region 4 (Chiapas, Mexico), 14 transects (1560 min, 13,000 m) were conducted in 1999 and 2001. At most sites in Oaxaca and Guerrero we found relatively intact habitats but greatly reduced numbers of both species and individuals of amphibians. Some of the stream courses near the Río Jalatengo, Oaxaca (Region 2) had been selectively logged, but in Veracruz we found widespread habitat alteration that prohibited resurveys of historical localities. In Chiapas (Region 4) transects occurred mostly in intact habitats, although coffee cultivation was present in some areas.

Overall, we found 39 of the approximately 100 species previously recorded from these regions, and these species represented 19–48% of the known fauna at a given region. We report 22 species that are missing from one or more of our study areas; many of these were endemic to these regions, have not been seen in many years, and may be extinct. We collected a total of 161 adult anurans on transects, and another 40 individuals off-transect. We found as many as 15 individuals and 7 species on any particular transect. We did not encounter nearly as many species or individuals as expected when compared to historical records from the same areas.

Based on the information on breeding phenologies presented in Caldwell (1974a) and Duellman (2001), we expected to encounter at least 16 of the 23 species of *Hyla* known from our sites (no breeding information was available for 7 species). We found only 9 species of *Hyla* during our surveys. We did not encounter either adults or tadpoles of 10 species that should have been present during this time: *Hyla altipotens*, *H. celata*, *H. cembra*, *H. cyanomma*, *H. dendroscarta*, *H. juanitae*, *H. pellita*, *H. pentheter*, *H. pinorum*, and *H. thorectes*.

In the Sierra Madre del Sur, Guerrero (Region 1), we captured 44 individuals of 16 amphibian species in 17 transects (1260 person-min; 8250 m), representing 44% of the 36 expected anuran species. Fourteen of 94 (15%) tadpoles collected from two areas (near Agua del Obispo, and near Carrizal del Bravo) were missing mouthparts, including tadpoles of *Ptychohyla erythromma*, *Rana sierramadrensis*, and those referrable to a species in the *Hyla bistincta* group (most likely *H. chyrses* or *H. mykter*). We also found one dead *Eleutherodactylus saltator* and one dying *Ptychohyla erythromma*, both of which were infected with chytrid fungus. Diagnostic exams by Dr. Allan Pessier confirmed that both frogs were heavily infected by *B. dendrobatidis*; and that this was the likely cause of death. We found tadpoles with missing mouthparts in both streams in which we found dead frogs.

On the Pacific Slope of the Sierra de Miahuatlan (Region 2) we captured 35 individuals of 6 anuran species in 8 transects (420 person-min; 1950 m), representing 19% of the 32 expected species. Of the 58 tadpoles collected, we found 9 (16%) *Ptychohyla leonhardschultzei* with missing mouthparts from north of San Gabriel Mixtepec and from near the Río Jalatengo. We did not find the endemic species *H. pentheter*, but a group of herpetologists who visited this region in 2002 found a metamorphosing individual from a locality just north of Candelaria Loxicha (J.J. Wiens, pers. comm.).

Along the North Slope of the Sierra de Juárez (Region 3) we captured 42 individuals of 11 species in 17 transects (1220 person-min; 7975 m), representing 44% of the 25 expected species. We found that 34 of 69 (49%) tadpoles collected from 6 different streams in this area were missing mouthparts, including individuals of *Hyla nephila*, *H. sabrina*, *H. cyclada*, and tadpoles that we tentatively refer to *Duellmanohyla ignicolor*.

On the Pacific Slope of the Sierra Madre de Chiapas (Region 4), we captured 142 individuals of 14 species in 13 transects (1560 person-min; 13,000 m), representing 48% of the 29 expected species. We found that 4 of 120 (3.3%) tadpoles collected were missing mouthparts (3 *Plectrohyla matudai* and 1 *Plectrohyla sagorum*; all collected at sites over 1200 m elevation).

In Regions 1–3 we found at most 7 species and 15 individuals for any particular transect, with 40% of transects producing no individuals. Captures were relatively low regardless of which habitat (aquatic, terrestrial) was surveyed (Table 1). There was no significant difference between average stream and average terrestrial transect capture rates (Table 1) for captures/m/person ($p < 0.893$). Caldwell (1974a) found significantly more species ($\bar{x} \pm \text{std} = 1.89 \pm 0.11$ vs. 1.2 ± 0.29; $p < 0.006$) and more individuals per survey (10.5 ± 2.5 vs. 3.1 ± 6.9; $p < 0.006$) in 1970 than we encountered in 2000 (Fig. 2). If we restrict the comparison to captures

Table 1
Survey effort among habitats during 2000 surveys

Transect type	Summary data	Mean capture rate ± standard deviation (No. captures/meter/person)
Stream, diurnal ($n = 13$)	640 min; 3550 m 31 adult caps	0.003 ± 0.003
Stream, nocturnal ($n = 24$)	1540 min; 7125 m 92 adult caps	0.002 ± 0.002
Trail, diurnal ($n = 5$)	720 min; 7500 m 33 adult caps	0.002 ± 0.002
Trail, nocturnal ($n = 1$)	60 min; 800 m 5 adult caps	0.0002

Capture rates of anurans for each transect type are expressed in terms of captures per meter of transect walked per person.

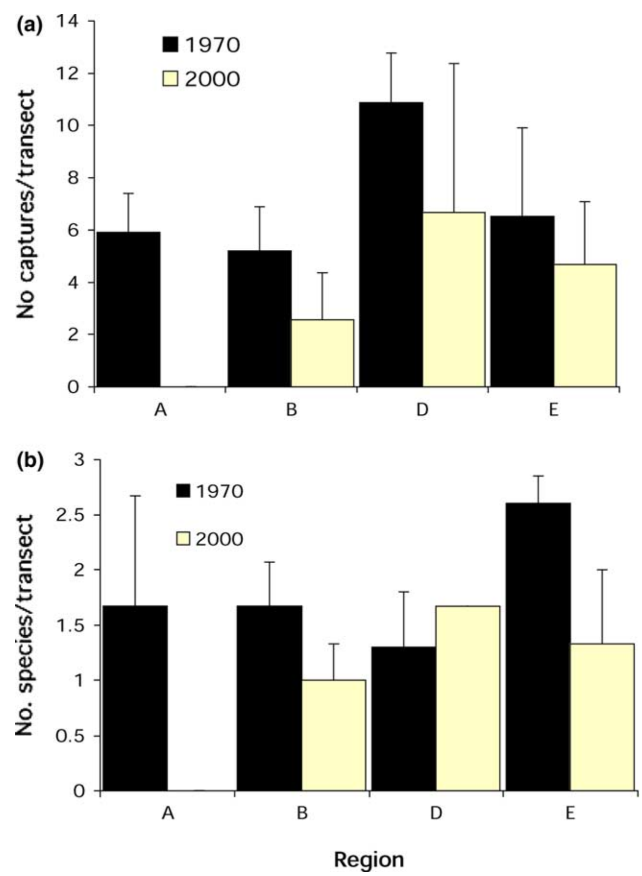

Fig. 2. Average number (±SD) of amphibian (a) captures and (b) species per transect in 1970 and 2000 for 4 of Caldwell's 5 regions. Area designations are those of Caldwell (1974a): Area $A = 0$–1.3 km N Cerro Pelón, Area $B = 1$–17 km S Vista Hermosa, Area $D = 20$–39 km N San Gabriel Mixtepec, and Area $E = 2$–15 km N San Gabriel Mixtepec. Data are only for nocturnal stream transects conducted during June–July of each year. We did not survey streams at night for any site in Region C during 2000, so we have removed that region from the figure.

Table 2
Total number of tadpoles collected per species, and incidence of loss of keratinized mouthparts

Species	No. lots	No. tadpoles	No. tadpoles with complete mouthparts	No. tadpoles with incomplete mouthparts (%)
Duellmanohyla ignicolor	4	13	2	11 (85)
Hyla cyclada	2	7	6	1 (14)
Hyla nephila	7	23	13	10 (43)
Hyla sabrina	7	17	8	9 (53)
Hyla pentheter	1	101	101	0 (0)
Hyla melanomma	2	13	4	9 (69)
Hyla sp. indet.	3	17	16	1 (6)
Hyla sumichrasti	17	107	104	0 (0)
Ptychohyla erythromma	1	1	0	1 (100)
Ptychohyla leonhardschultzi	4	11	3	8 (73)
Ptychohyla zophodes	2	13	6	7 (54)
Rana berlandieri	4	12	12	0 (0)
Rana forreri	1	1	1	0 (0)
Rana sierramadrensis	1	4	1	3 (75)
Rana zweifeli	1	8	8	0 (0)
Smilisca baudini	5	20	17	0 (0)
Sum	62	368	305	60 (19.67%)
Mean	3.647	21.647	17.941	3.53 (34.53%)
SD	3.968	31.672	32.252	4.303
Range	1–17	1–107	0–104	0–11 (0–100%)

from nocturnal stream transects conducted during the months of June and July, then there was a significant difference between the number of captures between 1970 and 2000 (Wilcoxon/Kruskal–Wallis rank sum tests, $X^2 = 4.94$, $p < 0.0263$), although not for the number of species (Wilcoxon/Kruskal–Wallis rank sum tests, $X^2 = 3.194$, $p < 0.0739$). Only Area B showed a significant difference between years, and did so for both the number of captures (Wilcoxon/Kruskal–Wallis rank sum tests, $X^2 = 4.438$, $p < 0.0351$) and species (Wilcoxon/Kruskal–Wallis rank sum tests, $X^2 = 4.357$, $p < 0.0368$).

Capture rate was highest between 500 and 1300 m elevation, although there was no significant relationship between capture rate and elevation for the number of anuran captures/m/person ($r^2 < 0.001$, $p > 0.893$) on all 43 transects, nor for the 24 nocturnal stream surveys alone ($r^2 = 0.014$, $p > 0.577$).

We collected 368 tadpoles on 23 of the 40 transects; 11 transects produced tadpoles that were missing some or all of their mouthparts, which can indicate infection by chytrid fungus (Lips, 1998, 1999; Fellers et al., 2001). Sixty tadpoles (19%) representing 9 hylid species and one species of *Rana* were collected with missing mouthparts (Table 2); these came from 9 different streams occurring at elevations above 800 m in our study regions.

Our efforts to conduct surveys in the Sierra Madre Oriental, in the State of Veracruz, were hampered by extensive habitat degradation. We could not positively locate historical collecting sites and streams, nor would we know whether population declines were due to habitat loss or other factors. We did conduct an interview with a longtime local resident and naturalist, and report his observations below.

4. Discussion

We found more anurans at lower elevations than we did at higher elevations. Even our highest capture rates were lower than that for similar habitats surveyed by KRL in Costa Rica and Panama (Lips, 1999, unpublished data). Capture rates for both terrestrial and riparian transects surveyed by KRL in central Panama are generally 0.1–0.5 captures/m which is 10–100 times greater than our Mexican surveys. In his extensive fieldwork throughout Mesoamerica during the 1950s–1970s, W.E. Duellman (pers. comm.) found capture rates to be similar between Mexico and similar habitats of Central America. We were unable to find many species that were common in surveys conducted during previous decades. In addition to widespread evidence of population declines and missing species, we found direct evidence of amphibian mortality in two regions and loss of keratinized mouthparts in tadpoles from all four regions. Duellman (2001, p. 816) found no missing mouthparts during an examination of tadpoles of 28 species collected in Mexico, Guatemala, Costa Rica, and Panama between 1960 and 1970.

Species that were not detected, or those found in reduced populations, were generally upland (>800 m elevation) populations of riparian species of hylids, centrolenids, and *Eleutherodactylus* (Table 3), as has been found in other areas in Latin America (Young et al., 2001; Lips et al., 2003b). We found moderate

Table 3
A conservative list of 31 anuran populations for which we found evidence of extinction or local extirpation in our four study regions

Region 1	Region 2	Region 3	Region 4
Agalychnis moreleti	*Agalychnis moreleti*	*Eleutherodactylus berkenbuschii*	*Agalychnis moreleti*
Eleutherodactylus guerreroensis	*Eleutherodactylus guerreroensis*	*Eleutherodactylus polymniae*	*Eleutherodactylus greggi*
Eleutherodactylus lineatus	*Eleutherodactylus lineatus*	*Eleutherodactylus werleri*	*Hyalinobatrachium fleischmanni*
Hyla hazelae	*Eleutherodactylus saltator*	*Hyla calivcollina*	*Plectrohyla guatemalensis*
Rana omiltemana	*Hyla altipotens*	*Hyla celata*	*Plectrohyla hartwegi*
	Hyla cembra	*Hyla cyanomma*	
	Hyla hazelae	*Hyla dendroscarta*	
	Hyla pellita	*Hyla echinata*	
	Hyla thorectes	*Hyalinobatrachium fleischmanni*	
	Hyla trux	*Smilisca cyanosticta*	
	Hyalinobatrachium fleischmanni		

Five species are apparently missing from several sites.

populations of various species of pool- and pond-breeding treefrogs (e.g., *Hyla smithi*) at several sites at lower elevations, but no amphibians were abundant at any site.

Some of our study sites have been repeatedly visited over several decades, so we were able to compile fairly complete species lists for most of our sites. Therefore, when we did not encounter a particular species, we concluded that abundances of those populations had decreased. A short-term visit may fail to detect the presence of an uncommon species, but many tadpoles of upland stream-breeding hylids are present in the streams throughout the year (Caldwell, 1974a) and should have been detected by our sampling. Absence of tadpoles of these species (e.g., *Hyla altipotens*, *H. celata*, *H. cembra*, *H. cyanomma*, *H. dendroscarta*, *H. juanitae*, *H. pellita*, *H. pentheter*, *H. pinorum*, and *H. thorectes*) most likely indicates large reductions in population abundance.

Except for Caldwell (1974a,b) extensive ecological research and field notes, most historical information from our study sites was derived from museum specimens and associated field notes made during short (days–weeks) visits by collectors; such data can only establish presence/absence of common species or those species or life-stages (e.g., some tadpoles) that are present year-round. Museum catalogues indicate which species were collected during the visit, but might not indicate those species seen or heard but not collected or those species not encountered. For example, J.A. Campbell visited Cerro Pelon multiple times during the 1980s on his way to Guatemala. During those visits he did not collect any specimens so did not record data in any field catalogue, but he noticed that the large, conspicuous frog *Hyla cyanomma* could no longer be found throughout the 1980s. We verified his estimated date by searching museum holdings for the last collected specimen, which in this case was 1980.

Our field surveys and museum studies of the frog fauna of montane southern Mexico suggest that most population declines occurred in this region in the mid- to late 1970s and early 1980s. Below we describe the changes in the frog fauna of each of these regions, paying special attention to the well-studied hylid fauna, and indicating the date when the last specimen was deposited in a musuem. Caldwell (1974a) usually encountered large populations and diverse communities of frogs at all of her sites. For example, she found 12 species in our Region 2. Some of these species were regularly encountered for 6 or more months of the year (e.g., *H. juanitae*, *H. pentheter*), or were occasionally abundant during that time. Furthermore, on multiple occasions in 1970, Caldwell observed choruses of at least 20 *H. juanitae*, 50 *H. pentheter*, and 20 *H. thorectes*. In 2000, we did not observe comparable population sizes or species richness at these same sites. We conclude that at least 5 species endemic to Regions 1 and 2 are missing and might be considered extinct, including *Hyla hazelae* (last collected in 1984), *H. thorectes* (last collected in 1984), *H. altipotens* and *H. pellita* (both last collected in the 1960s), and *Rana omiltemana* (last collected in 1978). Populations of at least 4 other species have declined and may be extirpated from these regions, including *Eleutherodactylus guerreroensis* and *Agalychnis moreleti* (both not collected since 1984), *E. lineatus, E. rugulosus*, and *Hyla trux*, *Hyalinobatrachium fleischmanni*.

Caldwell (1974a) found 16 species in our Region 3, and some of these species were either regularly encountered throughout the year (e.g., *H. cyclada*, *H. nephila*, *H. sabrina*) or were occasionally abundant. For example, Caldwell (1974a) recorded seeing more than 13 *H. celata* in a single day in June 1970, and 68 *H. cyanomma* on 10 May 1970. Our observations in 2000 do not even closely approximate the species richness and abundance that Caldwell (1974a) observed. Other collecting trips to the site by JRM (pers. obs.) and J.A. Campbell (pers. comm.) at various times in the 1990s also encountered a depauperate frog fauna relative to what Caldwell (1974a) observed. At least 6 species endemic to this site appear to be missing and might be considered extinct, including *Hyla calvicollina* and *H. celata* (both not collected since 1984), *H. cyanomma* (not collected since 1980), *H. dendroscarta* (not collected since 1974), *H. echinata* (last collected prior to 1962), and *Eleutherodactylus polymniae* (not collected since 1983). Populations of at least 5 other species have declined and may be extirpated from Region 3, including *Agalychnis moreleti*, *Smilisca cyanosticta*, *Eleutherodactylus berkenbuschii*, *E. werleri*, and *Hyalinobatrachium fleischmanni*. The case for the apparent extinction of *H. cyanomma* is particularly compelling. This was a common and conspicuous species, and herpetologists visiting streams on the upper slopes of Cerro Pelón routinely encountered tadpoles and adults (Caldwell, 1974a,b; R. Altig, pers. comm.; J.A. Campbell, pers. comm.; W.E. Duellman, pers. comm.) at all times of the year. Despite frequent visits by many herpetologists to these forested streams, adults of *H. cyanomma* have not been seen since 1978 and tadpoles were last seen in 1980 (J.A. Campbell, pers. comm.).

We estimate that at least 5 species have been extirpated from the El Triunfo Biosphere Reserve in the Sierra Madre de Chiapas. *Plectrohyla hartwegi* has not been collected since 1993 (Luna, 1997); *P. guatemalensis* has not been collected since 1944 (Luna, 1997); and *Eleutherodactylus greggi* and *Hyalinobatrachium fleischmanni* have not been seen since at least 1997. We found tadpoles of two species of *Plectrohyla* with missing mouthparts, indicating the likely presence of chytrid fungi in this region.

The taxonomic and ecological patterns of the declines and losses that we observed in Mexican frogs are similar to those described for amphibian population declines in Costa Rica and Panama (Pounds et al., 1997; Lips, 1998, 1999; Lips et al., 2003b); large, endemic, riparian species showed the greatest losses (e.g., *Eleutherodactylus rugulosus*, *E. guerreroensis*, *E. berkenbuschii*, *H. cyanommma*, *A. moreleti*, and *Rana omiltemana*). Populations of five more widespread species (*Agalychnis moreleti*, *Eleutherodactylus guerreroensis*, *E. lineatus*, *Hyla hazelae*, and *Hyalinobatrachium fleischmanni*), have been extirpated from multiple sites. Similarity in response among conspecific populations was also noted for the Costa Rican and Panamanian declines (Lips et al., 2003b). *Hyalinobatrachium fleischmanni* is one of the few species shared by our sites in Mexico and those sites in Costa Rica and Panama. Historical densities in Monteverde, Costa Rica were approximately 1 male per meter of stream transect, while today populations consist of a few males in several hundred meters in both Costa Rica (Pounds et al., 1997) and Mexico (pers. obs.).

B. dendrobatidis is currently present in each of the upland regions we surveyed, and was likely involved in population declines from these relatively undisturbed areas, although the role of this pathogen in population declines is not known. It is possible that factors other than habitat disturbance and chytrid fungi are involved, but we have no data to identify those factors. We found evidence of pollution and habitat loss in some parts of southern Mexico; one stream, the Río Zumpango, was obviously polluted with a strong chemical smell, dark brown water, and massive drifts of soapy bubbles. Despite these conditions, we found the second largest number of adult anurans in this stream, although many individuals had gross lesions on their livers. No other frogs from any other site had similar lesions. Amphibian declines in parts of Veracruz may be the result of deforestation. We interviewed Miguel Cerón, a naturalist in Cuautlapan, Veracruz, in the Sierra Madre Oriental who has been assisting herpetologists in the field ever since E.H. Taylor visited in the 1930s. Sr. Cerón confirmed that virtually all amphibians in the region have drastically decreased in abundance, and some have disappeared entirely. He mentioned that the hylid frog *Anotheca spinosa* had been extirpated from that region, the salamander *Lineatriton lineolus* had become less abundant than in previous decades, but that some species (e.g., *Smilica baudinii*, *Bufo marinus*, *B. valliceps*) were as common as ever. He attributed local declines to habitat alteration and pollution from a nearby cement factory.

Our data suggest that most amphibian population declines occurred in montane southern Mexico in the mid- to late 1970s and early 1980s. Habitat loss and degradation are likely culprits throughout much of the lowlands and urbanized areas, but chytrid fungus is likely involved in declines from protected upland areas. We propose southern Mexico as a spatio-temporal link between the widespread amphibian declines in western North America in the 1970s and early 1980s and more recent declines in Lower Central America in the mid-1980s and 1990s. If true, this would support the hypothesis that the chytrid is moving southward through Central America. Future research should examine biogeographic variation in fungal samples from throughout Latin America.

5. Summary

Our results suggest that as many as 27 anuran species have declined in upland areas of southern Mexico. In our Regions 1 and 2 alone, 11 species of frogs have been missing for over 15 years, and at least 9 populations of more widespread species are missing and may be locally extirpated. Most species now missing were last seen between 1978 and 1985. We found direct and indirect evidence of the fungus *B. dendrobatidis* at numerous sites, and suggest that it was likely involved anuran population declines in Guerrero, Oaxaca, and Chiapas, Mexico. These findings represent a spatio-temporal link between documented declines and extinctions in North America and Lower Central America.

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