Spatial Tests of the Pesticide Drift, Habitat Destruction, UV-B, and Climate-Change Hypotheses for California Amphibian Declines

CARLOS DAVIDSON,* H. BRADLEY SHAFFER,† AND MARK R. JENNINGS‡

*Department of Environmental Studies, California State University—Sacramento, 6000 J Street, Sacramento, CA 95819–6001, U.S.A., and Graduate Group in Ecology, University of California, Davis, CA 95616, U.S.A., email cdavidson@csus.edu
†Section of Evolution and Ecology, and Center for Population Biology, University of California, Davis, CA 95616, U.S.A.
‡Rana Resources, 39913 Sharon Avenue, Davis, CA 95616–9456, U.S.A., and Department of Herpetology, California Academy of Sciences, Golden Gate Park, San Francisco, CA 94118, U.S.A.

Abstract: *Wind-borne pesticides have long been suggested as a cause of amphibian declines in areas without obvious habitat destruction. In California, the transport and deposition of pesticides from the agriculturally intensive Central Valley to the adjacent Sierra Nevada is well documented, and pesticides have been found in the bodies of Sierra frogs. Pesticides are therefore a plausible cause of declines, but to date no direct links have been found between pesticides and actual amphibian population declines. Using a geographic information system, we constructed maps of the spatial pattern of declines for eight declining California amphibian taxa, and compared the observed patterns of decline to those predicted by hypotheses of wind-borne pesticides, habitat destruction, ultraviolet radiation, and climate change. In four species, we found a strong positive association between declines and the amount of upwind agricultural land use, suggesting that wind-borne pesticides may be an important factor in declines. For two other species, declines were strongly associated with local urban and agricultural land use, consistent with the habitat-destruction hypothesis. The patterns of decline were not consistent with either the ultraviolet radiation or climate-change hypotheses for any of the species we examined.*

Pruebas Espaciales de la Deriva de Pesticidas, Destrucción de Hábitat, UV-B e Hipótesis de Cambio Climático para la Declinación de Anfibios de California

Resumen: *Por mucho tiempo se ha sugerido que los pesticidas transportados por el viento son una causa de la declinación de anfibios en áreas sin destrucción de hábitat evidente. En California, el transporte y depósito de pesticidas provenientes del Valle Central, donde se practica la agricultura intensiva, hacia la Sierra Nevada adyacente está bien documentado y se han encontrado pesticidas en el cuerpo de ranas de la Sierra. Por lo tanto, los pesticidas son una causa verosímil de las declinaciones, pero a la fecha no se han encontrado relaciones directas entre los pesticidas y la declinación de anfibios. Construimos mapas de sistemas de información geográfica del patrón espacial de las declinaciones de ocho taxones de anfibios de California, y comparamos los patrones de declinación observados con los esperados por las hipótesis de pesticidas transportados por el viento, la destrucción del hábitat, la radiación ultravioleta y el cambio climático. En cuatro especies, encontramos una fuerte asociación positiva entre las declinaciones y la cantidad de tierras de uso agrícola en dirección contraria a los vientos, lo que sugiere que los pesticidas transportados por el viento pueden ser un factor importante en las declinaciones. Para otras dos especies, las declinaciones se asociaron contundentemente con el uso del suelo urbano y agrícola, lo cual es consistente con la hipótesis de la destrucción del hábitat. Los patrones de declinación no fueron consistentes con la hipótesis de la radiación ultravioleta ni la de cambio climático para ninguna de las especies examinadas.*

Introduction

During the last decade, amphibian declines have emerged as a key example of the global biodiversity crisis. Although a great deal of effort has been expended on determining the worldwide causes of amphibian declines, no consensus has yet emerged. One critical aspect of these declines is their geographic, or landscape-level, patterns. Over sufficiently large geographic areas, spatial patterns of amphibian decline can be used to evaluate hypothesized causes because most hypotheses implicitly make predictions of the expected spatial patterns of decline.

By developing spatially explicit predictions based on competing mechanisms, we used large-scale geographic patterns of decline to evaluate the importance of four putative causes of decline. Based on explicit assumptions, we generated spatial predictions for the hypotheses of habitat destruction, UV-B radiation, climate change, and pesticide drift, and we evaluated each hypothesis statistically by comparing predicted and observed spatial patterns of decline for eight species of California amphibians. The power of this strategy resides in its broad, species-wide approach that avoids reliance on one or a few study sites and its ability to simultaneously evaluate multiple hypotheses for causes of declines (for related geographical approaches to amphibian declines see Alexander & Eischeid [2001]; Carey et al. [2001]; and Middleton et al. [2001]).

California is a hotspot of amphibian decline, with many species experiencing sharp range contractions (e.g., Jennings 1988; Fellers & Drost 1993; Jennings & Hayes 1994; Drost & Fellers 1996; Fisher & Shaffer 1996). The state also has a strong record of historic, museum-based collections and ongoing field surveys, providing the broad baseline of data necessary for our analysis (Shaffer et al. 1998). Our study is based on presence/absence data for multiple locations. "Present" sites are those that are currently occupied by a species, and "absent" sites are those that were previously occupied (based on museum or other historic records) but are currently not occupied. "Declines" refers to sites, or proportions of sites, that are currently absent.

We developed our spatial analysis strategy while conducting a detailed study of declines of the federally threatened California red-legged frog (*Rana aurora draytonii*) (Davidson et al. 2001). Although that study successfully identified several potential causes of decline, any analysis of patterns of decline for a single species potentially suffers from two limitations. First, there is always the possibility that factors correlated with but not included in the study may be driving actual declines. Second, it may be that causal interpretations are correct for the single species but that other declining taxa are responding to a different set of factors. For our study of red-legged frogs, we explored the first possibility by dividing California into three ecoregions and analyzing the factors associated with declines within and between regions. In this way, we examined whether the factors associated with declines remained significant after unspecified regional factors were controlled for. A more powerful way to address the possibility of confounding factors (and that adopted here) is to analyze declines of multiple species with distinct ranges. It is unlikely that confounding factors that might have created a pattern of decline associated with one species would generate a similar pattern for a second or third species with a distinct range.

We analyzed eight species of California amphibians known to be declining based on federal or state listing status or analyses by Jennings and Hayes (1994). To be amenable to spatial analysis, a species must have a large geographic range in the state and reasonable numbers of both present and absent sites. Based on these criteria, we selected the following eight species to analyze: California tiger salamander (*Ambystoma californiense*), western spadefoot (*Spea hammondii*, sometimes referred to as *Scaphiopus hammondii*), arroyo toad (*Bufo californicus*), Yosemite toad (*B. canorus*), California red-legged frog (*Rana aurora draytonii*), Cascades frog (*R. cascadae*), foothill yellow-legged frog (*R. boylii*), and Sierra Nevada populations of the mountain yellow-legged frog (*R. muscosa*).

Six primary hypotheses have been proposed to explain amphibian declines: habitat destruction, pesticide drift, increased UV-B radiation, climate change, introduced exotic predators, and disease. We chose to analyze the first four hypotheses because we could generate clear and testable predictions for the spatial pattern of declines for each. In generating spatial predictions, our key underlying assumption is that increasing levels of an environmental stressor, be it completely anthropogenic (pesticides, habitat destruction) or human-mediated changes in the level of natural conditions (UV-B radiation, temperature), will increase the probability of declines. Our assumption is consistent with local adaptation as long as adaptation does not completely mitigate increased exposure to a stressor. Both empirical (Gross & Price 2000) and particularly theoretical (Kirkpatrick & Barton 1997) analyses of species' range limits support the view that adaptation only partially mitigates stressors and that species cannot infinitely adapt to environmental gradients. Instead, at the upper end of environmental gradients, population sizes decrease and the likelihood of decline increases.

If habitat destruction or modification associated with intensive human activities is contributing to amphibian declines, one would expect to see greater declines at sites with greater amounts of surrounding urban or agricultural land use than at those surrounded by wildlands. Such habitat effects could be due to direct habitat destruction or more indirect effects such as increased mortality from automobiles (Fahrig et al. 1995), increased predation by human commensals and pets (Crooks & Soulé 1999), pond modifications that favor exotic preda-

tors (Adams 1999), or habitat fragmentation (Marsh & Pearman 1997; Vos & Chardon 1998).

Wind-borne pesticides have long been suggested as a cause of amphibian declines (Carey & Bryant 1995; Stebbins & Cohen 1995; Drost & Fellers 1996; Lips 1998) in areas without obvious habitat destruction. California farmers used over 90 million kg of pesticide-active ingredients in 1998 alone (Department of Pesticide Regulation 1998). Transport and deposition of pesticides from the agriculturally intensive Central Valley of California to the adjacent Sierra Nevada is well documented (Zabik & Seiber 1993; Aston & Seiber 1997; McConnell et al. 1998; LeNoir et al. 1999), and pesticides have been found in the bodies of Sierra frogs (Cory et al. 1970; Datta et al. 1998). Low levels of pesticides can cause fatal immune suppression in amphibians (Taylor et al. 1999*a*). These studies indicate that pesticides, either alone or in combination with other stressors (Boone & Semlitsch 2001; Relyea & Mills 2001), are a plausible cause of declines but do not link pesticides and actual amphibian population declines. If wind-borne pesticides or other agrochemicals are a major factor in declines, one would expect greater declines at sites with greater amounts of agricultural land use upwind from the site.

The global warming and UV-B radiation hypotheses predict specific patterns of decline associated with elevation and latitude. Global warming is expected to shift species ranges poleward and up slope to higher elevations (Peters 1991; Parmesan 1996). If global warming were a major contributor to California amphibian declines, one would expect more declines in southern latitudes and fewer declines to the north. Similarly, more declines would be expected at lower elevations. Global warming may also affect frogs through changes in precipitation (Pounds & Crump 1994; Laurance 1996; Pounds et al. 1999), in which case one might expect to see proportionately greater declines at drier sites. Climatic data for 1900–1994 indicate that all California state climate divisions show an increase of 3° C per century in average daily temperature and a decrease of 20% per century in average precipitation (Karl et al. 1996). Under the UV-B hypothesis, we predict proportionately greater declines both at higher elevations and at more southerly latitudes where there is greater UV-B exposure (Blumthaler 1993; Cabrera et al. 1995; Madronich et al. 1995; Herman et al. 1999). Although there is a perception that amphibian declines have been concentrated at high elevations, and thus that UV-B radiation is a potential causal agent, to date the pattern has been quantified (with mixed results) for only a single species (Davidson et al. 2001).

Methods

We used maps produced by Jennings and Hayes (1994) to document the spatial patterns of decline for eight Cal-

ifornia amphibians. The maps are based on verified museum records and recent surveys, and provide the most comprehensive evaluation of California declines available for a single point in time. We used Arc/Info geographic information system (GIS) (versions 7.1.1. and 7.2.1) to digitize eight of these maps and register the maps to an Albers projection digital county map of California. This produced a spatial data set of 1491 sites, spanning the entire state. (See Table 1 for number of sites per species and Fig. 1 for a map of sites for the four ranid frog species.) We used U.S. Geologic Survey 1:250,000–scale digital elevation models for California to derive elevation for all sites, and we estimated the 60-year (1900–1960) average annual precipitation for each site based on a Teale Data Center digital precipitation map of California. Latitude for each location was determined directly from the coordinates for the site. To assess the contribution of habitat destruction to declines, we measured the percentage of urban and agricultural land use in a 5-km radius surrounding each site based on digital 1:250,000–scale land use/land cover maps from the U.S. Geological Survey (USGS).

We used percentage of upwind agricultural land use as a proxy for the intensity of wind-borne agrochemicals that sites experience. For this analysis, we first estimated predominant summer wind direction for each site from streamline wind maps for California and wind-direction data from weather stations (Hayes et al. 1984). We used summer wind patterns because analysis of regional wind patterns in the San Francisco Bay Area, South Coast, Sacramento, and San Joaquin regions indicates that the predominant summer wind pattern in all regions is also the predominant annual wind direction (Hayes et al. 1984). Summer and spring, which have similar wind patterns, are also when roughly two-thirds of California agricultural pesticides are applied (Department of Pesticide Regulation 1990, 1994). To define the area we considered upwind from a site, we used GIS to construct an "upwind triangle," 22.5° (1 compass sector, where each sector equals one of the 16 standard compass directions) wide, 100 km long, and facing upwind (Fig. 2). We also constructed 33.75°– and 45°–wide triangles (1.5 and 2 compass sectors) for comparison. We used USGS digital 1:250,000–scale land use/land cover maps to calculate the percentage of the total area within an upwind triangle that was agricultural land. We call this measurement "percent upwind agricultural land use" or "upwind agriculture" for short.

To assess the joint effect of proximity to agricultural land use and the amount of agricultural land upwind from a site, we calculated an "upwind agricultural index." (Fig. 2). We divided all agricultural land within the upwind triangle into patches of ≤ 10 km^2. For each site we calculated the upwind agricultural index as $\Sigma\,(a_i/d_i)$, where a_i is the area of the ith patch of agricultural land within the upwind triangle, d_i is the distance from the

Table 1. Comparison of mean site characteristics for sites in which amphibians were present (present sites) versus sites in which amphibians were absent (absent sites).

Characteristic	Ambystoma californiense	Spea hammondii	Bufo californicus	Bufo canorus	Rana aurora draytonii	Rana boylii	Rana cascadae	Rana muscosa
Sites								
total	163	167	78	55	279	424	70	255
present	107	81	35	26	88	196	24	43
absent	56	86	43	29	191	228	46	212
percent absent	34	51	55	52	68	54	66	83
Latitude								
present sites	37.02	35.83	33.72	37.75	36.43	39.35	41.21	37.91
absent sites	37.42	34.91	33.61	37.88	36.11	37.68	40.38	37.87
p[a]	0.02	0.00	0.31	0.29	0.13	0.00	0.00	0.73
Elevation (m)								
present sites	266	327	723	2834	276	579	1651	2676
absent sites	103	227	553	2420	445	521	1542	2253
p	0.00	0.00	0.04	0.10	0.00	0.12	0.54	0.00
Upwind ag[b]								
present sites	0.28	0.38	0.06	0.05	0.04	0.12	0.05	0.11
absent sites	0.29	0.24	0.04	0.09	0.22	0.32	0.21	0.22
p	0.38	0.00	0.03	0.39	0.00	0.00	0.00	0.02
Surrounding urban[c]								
present sites	0.07	0.04	0.01	0.00	0.07	0.04	0.01	0.00
absent sites	0.22	0.23	0.13	0.01	0.10	0.05	0.01	0.00
p	0.00	0.00	0.00	0.12	0.02	0.00	0.04	0.16
Surrounding ag[d]								
present sites	0.30	0.27	0.04	0.00	0.09	0.04	0.00	0.00
absent sites	0.35	0.32	0.11	0.00	0.13	0.15	0.00	0.00
p	0.39	0.52	0.01	0.34	0.12	0.00	0.20	0.19
Precipitation (cm)								
present sites	16.3	15.4	19.5	44.3	23.2	48.1	54.5	46.2
absent sites	16.5	14.8	15.8	44.3	21.0	27.9	56.5	44.9
p	0.77	0.08	0.00	0.73	0.11	0.00	0.65	0.58
Random ag[e]								
present sites	0.24	0.22	0.07	0.03	0.14	0.13	0.04	0.05
absent sites	0.34	0.16	0.05	0.07	0.19	0.21	0.09	0.09
p	0.08	0.19	0.30	0.30	0.10	0.00	0.04	0.34
Downwind ag[f]								
present sites	0.24	0.12	0.10	0.01	0.20	0.10	0.03	0.01
absent sites	0.35	0.17	0.07	0.01	0.19	0.18	0.04	0.02
p	0.01	0.05	0.02	0.10	0.13	0.00	0.29	0.22
Upwind ag index[g]								
present sites	10,867	14,148	2,494	996	1,739	3,879	1,790	2,463
absent sites	12,001	9,965	2,375	1,946	7,712	10,734	4,776	5,464
p	0.60	0.00	0.29	0.39	0.00	0.00	0.00	0.01

[a] *The* p *value for Mann-Whitney test of difference of means between present and absent sites.*
[b] *Percentage of agricultural (ag) land use upwind of a site.*
[c] *Percentage of urban land use in a 5-km radius surrounding a site.*
[d] *Percentage of agricultural (ag) land use in a 5-km radius surrounding a site.*
[e] *Percentage of agricultural (ag) land use in a triangle oriented in a random direction.*
[f] *Percentage of agricultural (ag) land use in a triangle oriented in a downwind direction.*
[g] *An index that combines area and distance to agricultural (ag) land use upwind from a site.*

centroid of the *i*th patch to the amphibian site, and the summation is across all patches within an upwind triangle. The index is thus an inverse-distance, weighted measure of the area of agricultural land upwind from a site. In interpreting both the percentage of upwind agricultural land use and the upwind agriculture index, we focused on pesticides because of their toxicity and documented long-range transport. However, a pattern of declines statistically associated with upwind agricultural land use could be driven by other wind-blown agricultural substances that negatively affect frogs (e.g., fertilizers; Marco et al. 1999).

We used univariate, nonparametric Mann-Whitney rank tests (Sokal & Rohlf 1995) to evaluate differences in the mean value of characteristics for present and absent sites. We also analyzed key variables as categorical variables and plotted them to assess whether there was a consistent quantitative relationship between changes in
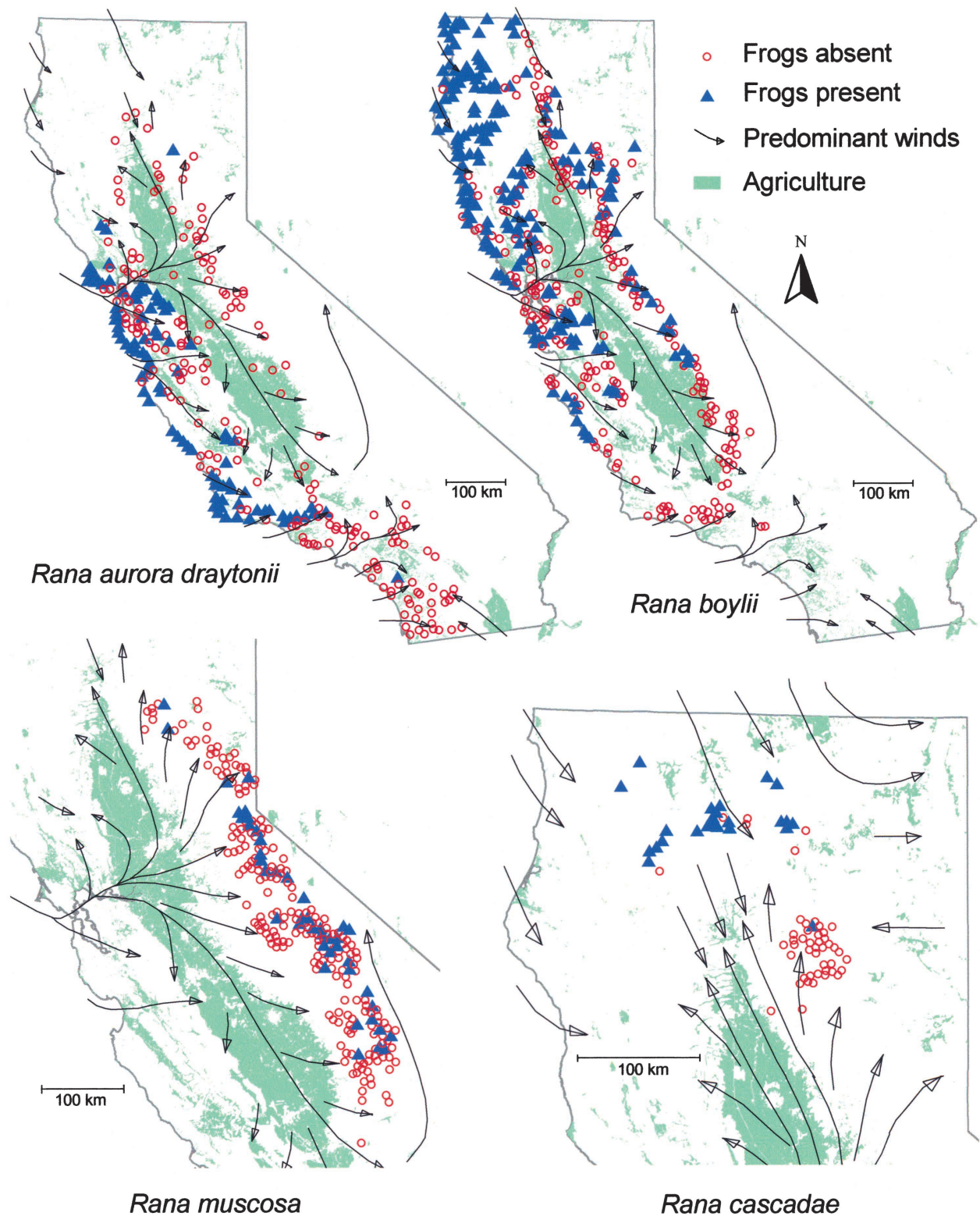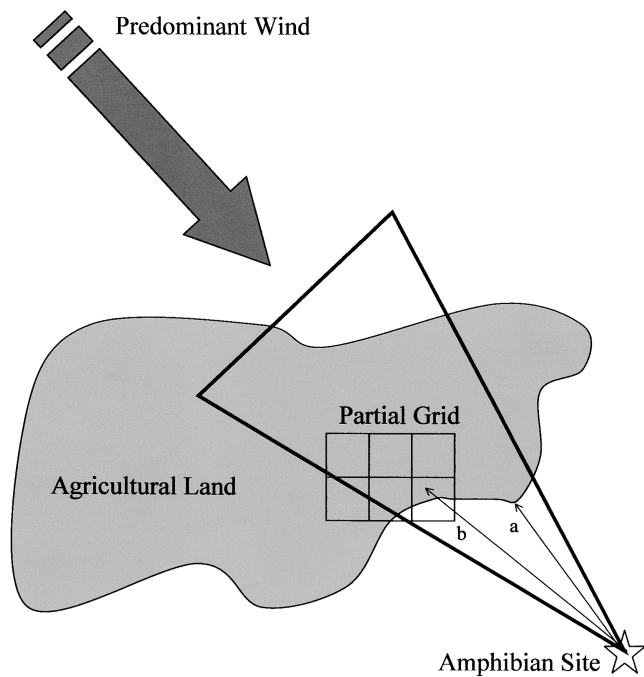
Figure 1. Spatial patterns of decline for four California ranid frogs, with the location and current population status for frog sites used in our analysis. Also shown are the distribution of agricultural lands based on U.S. Geologic Survey land use/land cover maps, and key predominant wind directions based on California Air Resources Board streamline wind maps.

Figure 2. Illustration of measurement of upwind agricultural land use. For each amphibian site, we drew a 22.5°, 100 km long "upwind triangle" facing into the direction of the predominant wind. Within the triangle, we measured upwind distance to the nearest agricultural land use (arrow a) and the percentage of the total area of the triangle consisting of agricultural land. An "upwind agricultural index" was calculated by dividing all agricultural land within the upwind triangle into patches 10 km² or less (based on the partially shown grid). For each patch we calculated patch area divided by the distance from the patch centroid to the amphibian site (arrow b). The index value was then the sum of these calculations across all patches within the upwind triangle.

the variable and the proportion of sites with declines. We used chi-square tests to evaluate the significance of the relationship between population status and each categorical variable and multiple logistic regression to evaluate the multivariate relationship between declines and geographic, precipitation, elevational, and land-use variables (Hosmer & Lemeshow 1989). For each species, we built a full model with all the variables and then one by one removed variables with statistically insignificant coefficients to derive a reduced model with only significant variables.

Our analyses rely on both the spatial accuracy of the maps and the accuracy of the characterization of site population status (present or absent). We performed several analyses to assess the accuracy of Jennings and Hayes's species maps and the sensitivity of our results to possible errors in site spatial location and population status. To check spatial accuracy, we created maps from

original museum records and the published literature (for methods see Davidson et al. 2001) for *R. a. draytonii* and *R. cascadae* and compared these maps to those produced by Jennings and Hayes (1994). We located the same sites on both maps and calculated the average spatial location error, assuming the original records were accurate. We chose these two species as exemplars both of species with large ranges within California and correspondingly small-scale maps (*R. a. draytonii*) and of species with small ranges in the state and therefore large-scale maps (*R. cascadae*), and we quantified spatial accuracy at both scales. To assess the robustness of our results to errors in spatial accuracy, we created a data set in which we randomly moved all sites in our original data set in a random direction and for a random distance ranging from zero to two times the estimated average spatial-location error. We used either the estimated *R. a. draytonii* or *R. cascadae* spatial-location error, depending on the scale of the original map. The reduced logistic regression models were then run on this new data set and the results compared with those on the original data. To assess the robustness of our results to possible errors in site population status (present/absent), we randomly switched status at approximately 10% of sites for each species and reran the reduced logistic regression models. We repeated this process 10 times and examined the number of times a variable in the original models remained statistically significant.

Results

The percentage of formerly occupied sites that are now absent was high for all eight species, ranging from 33% absent (*A. californiense*) to 83% absent (*R. muscosa*). In general, the results of the univariate (Table 1) and multivariate analyses were similar. We focused on the eight multiple logistic regression analyses to discern across-species patterns of variables associated with declines (Table 2). In all eight logistic regression models the likelihood-ratio test for the overall model was significant, the Hosmer-Lemeshow goodness-of-fit test (Hosmer & Lemeshow 1989) indicated that the data fit the model, and the models correctly classified population status for 63% (*B. canorus*) to 83% (*R. cascadae* and *R. muscosa*) of all sites. Use of upwind agriculture variables based on 33.75°- or 45°-wide triangles produced regression coefficients and significance levels nearly identical to those based on the triangle 22.5° wide; therefore, we report only results based on the 22.5° upwind triangle. Pairwise correlations of variables within species were generally below 0.5, so we discounted high colinearity in interpreting our mulitvariate results.

In the multivariate regression models, urbanization was a significant negative variable (that is, with increasing surrounding urban land use, sites were less likely to

Table 2. Logistic regression models of site population status for eight California amphibians.[a]

Variable	Ambystoma californiense	Spea hammondii	Bufo californicus	Bufo canorus	Rana aurora draytonii	Rana boylii	Rana cascadae	Rana muscosa
Latitude					+	+		
Elevation	+			+	−			+
Upwind ag[b]			+		−	−	−	−
Surrounding urban[c]	−	−	−		−			
Surrounding ag[d]		−	−			−		
Precipitation						+		
Goodness of fit[e]	0.85	0.23	0.57	0.16	0.13	0.21	0.99	0.67
Accuracy[f]	0.71	0.70	0.69	0.63	0.78	0.72	0.83	0.83

[a] *The model for each species is read down the column. The plus (+) and minus (−) signs indicate a significant coefficient for this variable and the sign (positive or negative) of the coefficient. In all models the dependent variable is site status (present, 1; absent, 0).*
[b] *Percentage of agricultural (ag) land use upwind of a site.*
[c] *Percentage of urban land use in a 5-km radius surrounding a site.*
[d] *Percentage of agricultural (ag) land use in a 5-km radius surrounding a site.*
[e] *The* p *value for the Hosmer-Lemeshow test.*
[f] *Proportion of sites correctly classified as present or absent sites.*

be a present site) for the salamander *A. californiense* and the anurans *S. hammondii*, *B. californicus*, and *R. a. draytonii*. When viewed in terms of mean urbanization at present and absent sites, the effect of surrounding urbanization was particularly striking for the first three of these species, with absent sites having 3 (*A. californiense*) to 13 (*B. californicus*) times greater local urbanization than sites where the species remained present (Fig. 3). The effect, although statistically significant, was much less pronounced for *R. a. draytonii*, whose absent sites on average were only about 50% more intensely urbanized than the present sites (Fig. 3). Surrounding agricultural land use in a 5-km radius was a significant negative variable for *S. hammondii*, *B. californicus*, and *R. boylii*.

Latitude was a significant positive variable for *R. a. draytonii*, and latitude and precipitation were positive variables for *R. boylii*. The model for *R. cascadae* was complicated by the fact that both latitude and percentage of upwind agriculture were each highly significant in individual univariate tests, yet the two variables were fairly highly correlated for this species (Pearson correlation, 0.64). There was a relatively small latitudinal difference (40 km) between the Lassen area, where the species has largely disappeared, and the Trinity Alps area, where the species is still common. There was also a strong, stepwise gradient in declines for percentage of upwind agriculture (Fig. 4) and no similar gradient for latitude. We therefore modeled declines for *R. cascadae* with the upwind agriculture variable and not latitude.

All four ranid frog species (*R. a. draytonii*, *R. boylii*, *R. cascadae*, and *R. muscosa*) showed a strong, statistically significant pattern of decline with greater amounts of upwind agriculture (Table 1; Fig. 1). In the univariate comparisons (Table 1; Fig. 3), the magnitude of this effect was large, with mean upwind agriculture for absent sites ranging from 2.0 (*R. muscosa*) to 4.2 (*R. cascadae*)

times greater than for present sites. The same trend was present for the montane toad *B. canorus*, although the difference in means (9% upwind agriculture for absent sites, 5% for present sites) was not statistically significant (Fig. 3), and upwind agriculture was not significant in the multivariate regression model for this species (Table 2). Similarly, the composite area and proximity agriculture index was greater for all five species at absent sites than present sites, and the difference was significant for the four ranid frog species but not for *B. canorus*. All five species showed a dose-response-like relationship with a gradient of greater declines with greater amounts of upwind agriculture (Fig. 4), and this relationship was significant for *R. a. draytonii*, *R. boylii*, and *R. cascadae*.

Elevation showed an unexpected pattern that was not consistent with our predictions for the UV-B hypothesis. For seven of eight species (all except *R. a. draytonii*; Fig. 3) the mean elevation of present sites was greater than that of absent sites, implying that within a species' range, declines have been concentrated at lower-elevation localities. In the logistic regression models for three species (*A. californiense*, *B. canorus*, *R. muscosa*), elevation was a significant positive variable (that is, the likelihood of a site being a present site increased with elevation).

There was only one significant interaction term in the reduced regression models: a negative term for latitude × urbanization for *R. a. draytonii*. That is, the negative effect of urbanization on the likelihood of a site being present was stronger at lower latitudes. The interaction term is consistent with the observation that the species is almost entirely absent near southern California urban areas, whereas it persists in the heavily urbanized San Francisco Bay Area.

Our results were reasonably robust to possible errors in spatial accuracy of the maps and site population status. The average spatial accuracy error for matched loca-
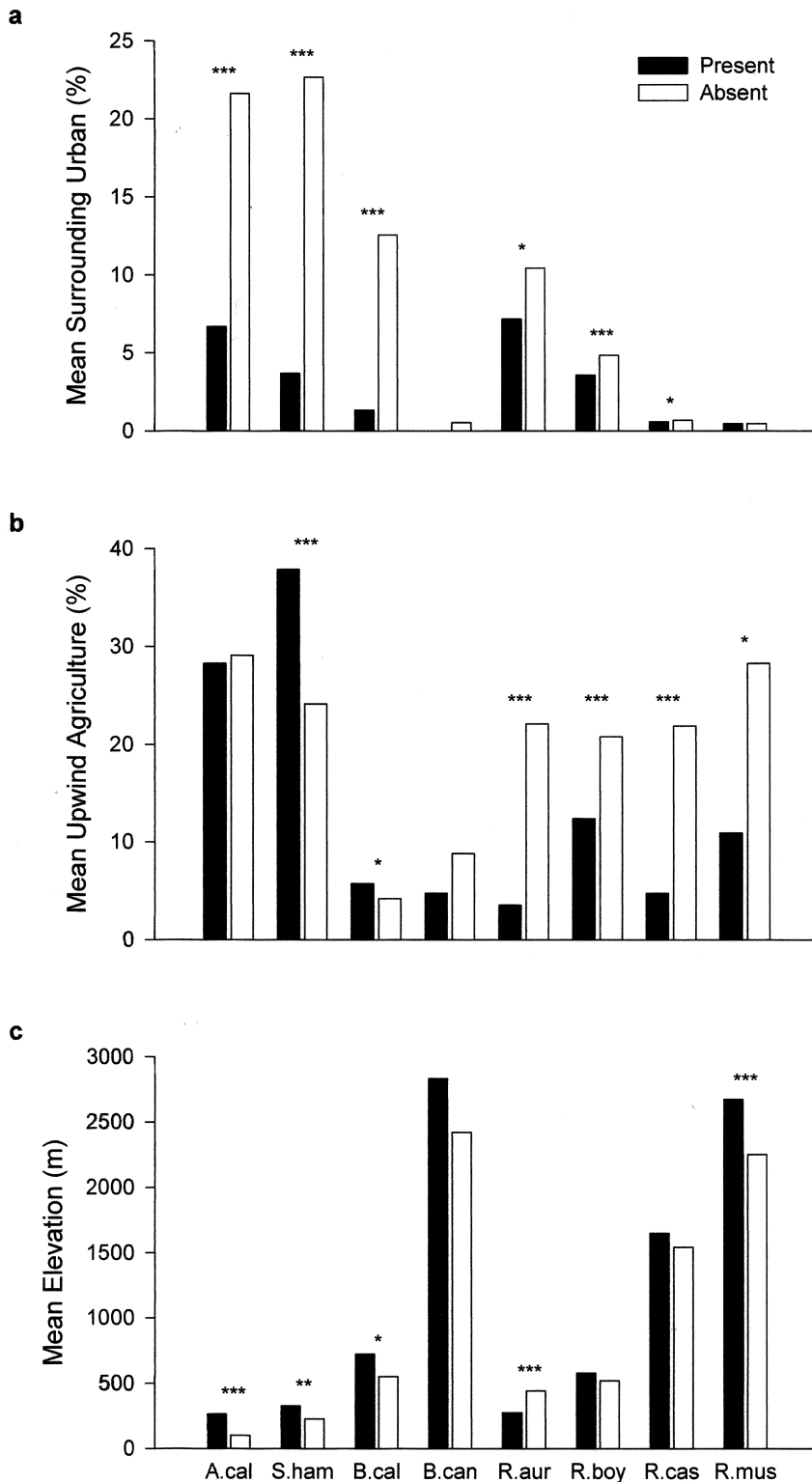
Figure 3. Univariate analysis of mean attributes of present and absent sites for eight species of declining amphibians. On the x-axis are species names abbreviations (e.g., A. cal is Ambystoma californiense). Mean values for present and absent sites are shown for three site attributes: (a) percent urban land use in a 5-km radius surrounding a site; (b) percent upwind agricultural land use in a 100-km triangle; and (c) elevation. Asterisks indicate p values for Mann-Whitney nonparametric tests of the difference of means between present and absent sites: *p < 0.05, **p < 0.01, and ***p < 0.001.

tions on the maps we derived from Jennings and Hayes and the maps we created from original records was 7.71 km for *R. a. draytonii* and 4.4 km for *R. cascadae*. The logistic regression models for all species were fairly robust to these levels of possible spatial-accuracy error.

The one exception was the *B. californicus* model, in which none of the variables were significant when run on the random-location error data set (surrounding urban, $p = 0.066$; surrounding agriculture, $p = 0.107$; upwind agriculture, $p = 0.27$). Five variables in specific
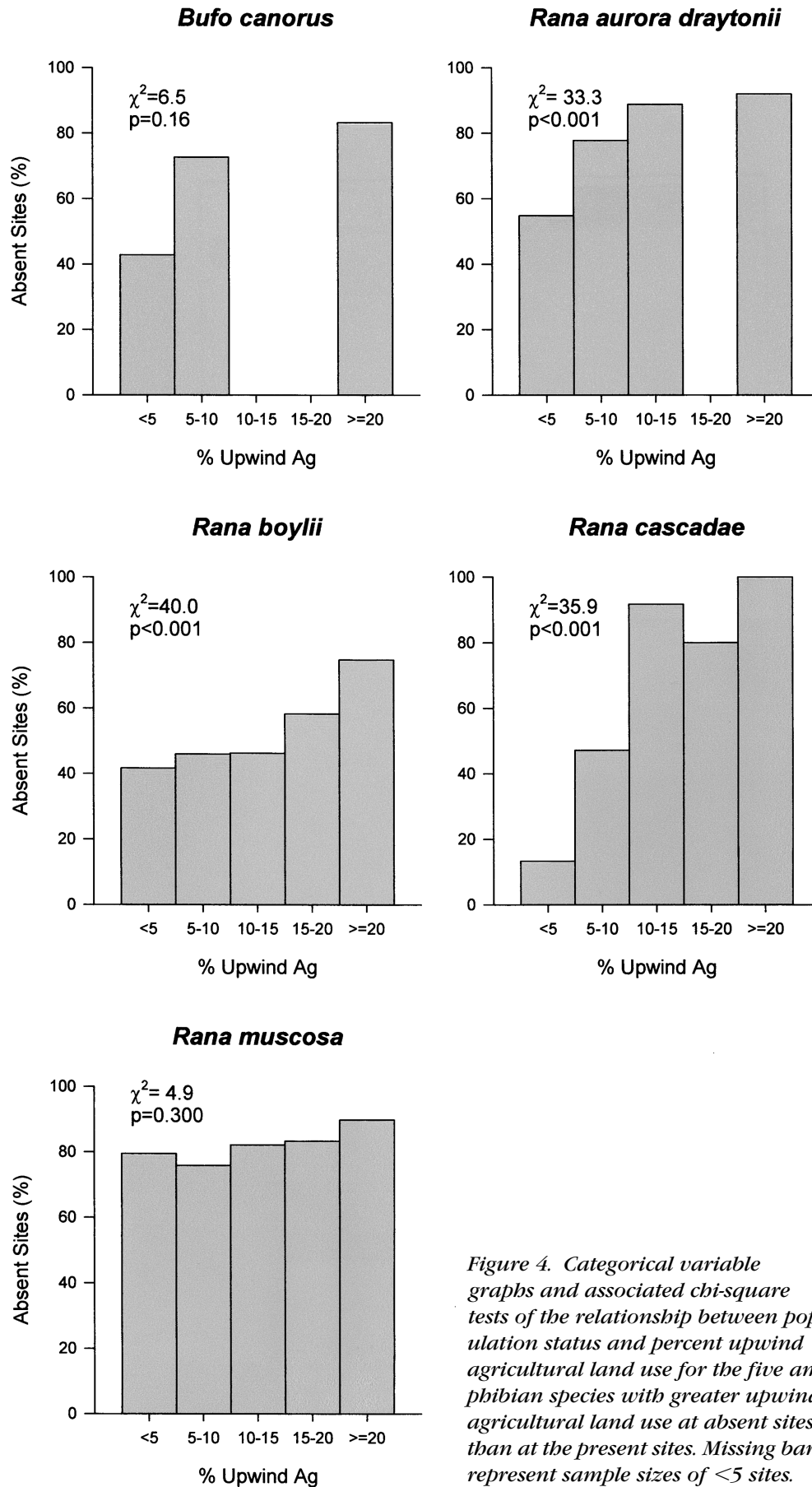
Bufo canorus

$\chi^2 = 6.5$
p=0.16

Absent Sites (%)

% Upwind Ag

<5   5-10   10-15   15-20   >=20

Rana aurora draytonii

$\chi^2 = 33.3$
p<0.001

Absent Sites (%)

% Upwind Ag

<5   5-10   10-15   15-20   >=20

Rana boylii

$\chi^2 = 40.0$
p<0.001

Absent Sites (%)

% Upwind Ag

<5   5-10   10-15   15-20   >=20

Rana cascadae

$\chi^2 = 35.9$
p<0.001

% Upwind Ag

<5   5-10   10-15   15-20   >=20

Rana muscosa

$\chi^2 = 4.9$
p=0.300

Absent Sites (%)

% Upwind Ag

<5   5-10   10-15   15-20   >=20

Figure 4. Categorical variable graphs and associated chi-square tests of the relationship between population status and percent upwind agricultural land use for the five amphibian species with greater upwind agricultural land use at absent sites than at the present sites. Missing bars represent sample sizes of <5 sites.

Table 3.   Support for hypotheses of causes of decline for eight California amphibians.[a]

Species	Habitat destruction	Climate change	UV-B radiation	Pesticides
Ambystoma californiense	×			
Spea hammondii	×			
Bufo californicus	×			
Bufo canorus				×[b]
Rana aurora draytonii	×[c]		/[d]	×
Rana boylii	×[c]	/[e]	/[f]	×
Rana cascadae				×
Rana muscosa				×

[a] An × indicates a spatial pattern of decline for the species consistent with the hypothesis; a slash (/) indicates a pattern that is only partially consistent with the hypothesis.
[b] B. canorus *shows greater declines with greater upwind agricultural land use, but the relationship is not statistically significant.*
[c] Habitat destruction is less of a factor for these two species than for the first two species in the table.
[d] Declines are positively associated with elevation, consistent with the UV-B hypothesis, but the predicted north-to-south gradient of increasing declines is not met.
[e] Declines are negatively associated with latitude and precipitation, consistent with the climate-change hypothesis, but the predicted increase in declines with higher elevation is not met.
[f] There is a north-to-south gradient of declines, consistent with the hypothesis, but the predicted increase in declines with higher elevation is not met.

models were sensitive to possible population status errors: urbanization for *A. californiense*, precipitation for *R. boylii*, and percent upwind agriculture for *B. californicus* and *R. muscosa*.

Discussion

The spatial analysis of patterns of decline is a powerful and rapid method for assessing potential factors that may have contributed to the widespread disappearance of many California amphibians. This strategy complements experimental studies of specific mechanisms; together, they contribute to establishing more certain cause-and-effect relationships between mechanisms and actual declines. In our study, we addressed predictions generated by four widely cited hypotheses for amphibian declines (summary in Table 3).

Habitat Destruction

We drew two primary conclusions from our analyses of habitat destruction as measured by surrounding urban and agricultural land use. First, for all eight species the amount of urban and agricultural land use was greater for absent than present sites. This confirms what we already know: both intensive local urbanization and agriculture tend to be inconsistent with the environmental requirements of amphibians. Overall, the message from these results is clear: in areas where habitats have been greatly modified by human activities, amphibian populations have declined accordingly. Some species still persist in highly modified habitats (for example, *A. californiense*), but all species suffer when their habitat is urbanized or converted to intensive agriculture.

Second, the importance of habitat destruction varies across species because of species-specific differences in both the amount of habitat destruction experienced and sensitivity to habitat destruction. For the two lowland species (*A. californiense* and *S. hammondii*) declines were strongly associated with surrounding urban land use and to a lesser extent with surrounding agricultural land use. These species are vernal-pool specialists (Stebbins 1951; Trenham et al. 2000; Shaffer & Trenham 2001), and this habitat has suffered extreme levels of destruction in California due to both urbanization and conversion to agriculture. Declines for *B. californicus* also show a strong association with surrounding urbanization in our analysis. We suspect, however (but cannot quantify), that declines are probably related to other forms of habitat alteration, such as recreational impacts on streams, modifications of flow regimes, and gravel mining, rather than directly to urbanization. The declines of the five remaining species are more puzzling because gross habitat destruction appears to be much less of a factor or not a factor at all. For the two low- to mid-elevation species (*R. a. draytonii* and *R. boylii*), habitat destruction appeared to be a contributing factor in declines, whereas for the three exclusively montane species, (*B. canorus*, *R. cascadae*, and *R. muscosa*) surrounding urbanization or agriculture has not played a role in declines.

Wind-borne Agrochemicals

The association of declines with the amount of upwind agricultural land use was striking for five of the six species for which habitat alteration was not a clear factor (Fig. 3), and it was a significant factor in the multivariate models for four of these taxa (Table 2). These results represent, at a minimum, three independent tests of the upwind agriculture pattern. The historic ranges of *R. a. draytonii* and *R. boylii* were roughly two-thirds overlapping (Fig. 1) and therefore could be considered a test for

a single geographical area. There was virtually no overlap, however, between the ranges of these two species and the ranges of *R. cascadae* and *R. muscosa* (Fig. 1). It is intriguing that declines were strongly associated with upwind agriculture for all four ranid species, suggesting that this group may be particularly sensitive to agrochemicals. Whether this is coincidental or significant is difficult to determine with only eight species, although the odds of getting this taxonomic pattern by chance alone are low ($p = 0.014$).

The strong association of decline for four species with the amount of upwind agricultural land use was not just a reflection of habitat alteration due to agriculture. Although the amount of upwind agricultural land use was associated with declines, the amount of agricultural land use in a 5-km radius surrounding a site was generally not associated with declines, except in the case of *R. boylii* (Table 2). Similarly, variables for agricultural land use in a downwind or random direction were not significant when added to the logistic regression models for these species, suggesting that the association reflects windborne agrochemicals rather than some generalized, nondirectional influence of agriculture. Additional evidence for the importance of upwind agriculture across species comes from the categorical variable analysis (Fig. 4), which demonstrated clear trends of increasing declines with increasing amounts of upwind agriculture. Of the factors we were able to examine, upwind agriculture was the strongest single factor explaining California declines for amphibian taxa in which declines were not driven primarily by overt habitat destruction.

UV-B Radiation

Our observed patterns of species decline are inconsistent with the predictions of the UV-B hypothesis. For all eight species pooled together, there were greater declines at higher elevations (above 1500 m, 72% of sites were absent sites, vs. 56% below 1500 m), which is consistent with reports that amphibian declines are concentrated at higher elevations (Wake 1991; Carey et al. 1999). However, this pattern was driven by greater overall declines in high-elevation species (mainly *R. muscosa*), rather than trends within species. When viewed individually, seven of eight species had a higher mean elevation at present sites than at absent sites (Fig. 3). This pattern is exactly opposite to the predictions of the UV-B hypothesis of greater declines at higher elevations. Results of an independent analysis of historical patterns of decline for the salamander *A. californiense* (Fisher & Shaffer 1996) shows this same elevational pattern.

Two species showed patterns partly consistent with the UV-B hypothesis. *Rana a. draytonii* had a clear gradient of increasing declines with elevation (as predicted by the UV-B hypothesis), but did not display the predicted north-to-south gradient (Davidson et al. 2001).

Rana boylii had the predicted north-to-south gradient of increasing declines (which could also be consistent with climate change) but had greater declines at lower elevations, opposite of that predicted by the UV-B hypothesis. Although UV-B exposure is affected by environmental variables such as canopy cover and dissolved organic content in water, as well as elevation and latitude, it is unlikely these factors would produce a systematic pattern of greater exposure at lower elevations. It is still possible that increased UV-B radiation may be adversely affecting amphibians, and experimental work on the biological effects of UV-B exposure needs to be evaluated on a species-by-species basis. However, under the assumptions of our analysis, the patterns of observed declines are not consistent with UV-B as a primary cause of amphibian declines in California.

Climate Change

Only *S. hammondii* had both significantly greater declines in the south and at lower elevations, as we predicted under the climate-change hypothesis. But neither variable was significant in the logistic regression model for the species (Table 2), and neither variable showed a gradient pattern in the univariate categorical variable analysis (data not shown). Declines for *S. hammondii* appeared to be more related to habitat degradation (Table 1; Fig. 3). The spatial pattern of decline for *R. boylii* was the most suggestive of an influence of climate change, with a strong gradient of increasing declines to the south and at drier sites. Because site latitude and precipitation are correlated for this species (0.58), we were unable to separate these two factors. Overall, our multispecies analysis did not implicate climate change, including precipitation effects, as a primary cause of amphibian declines in California. Although climate change may contribute to declines in more subtle or complex ways (e.g., Pounds et al. 1999; Kiesecker et al. 2001), we did not find large-scale spatial patterns of decline consistent with climate change, as have been found for other taxa (e.g., Parmesan 1996).

Other Potential Factors

We were unable to analyze the spatial implications of two other important hypotheses for declines: disease (for a review see Carey et al. 1999) and introduced species (e.g., Hayes & Jennings 1986; Fisher & Shaffer 1996; Kupferberg 1997; Adams 1999; Lawler et al. 1999; Knapp & Matthews 2000; Kiesecker et al. 2001b). Not enough is known about the biology of disease agents such as chytrid fungus (Berger et al. 1998; Taylor et al. 1999b), *Saprolegnia* (Blaustein et al. 1994; Kiesecker & Blaustein 1995), or the iridovirus (Cunningham et al. 1996) to generate testable spatial predictions. We did not have detailed site-by-site data on introduced species (primarily fish and bullfrogs [*R. catesbeiana*]) that

would allow us to include this factor in the current analysis. Disease and introduced exotic species undoubtedly both play a role in the decline of many California amphibians. For example, Bradford (1989), Bradford et al. (1994), Knapp and Matthews (2000), and V. Vredenberg (unpublished data) found that introduced trout have contributed to the declines of *R. muscosa*. In most cases, however, there have also been declines at sites without introduced fish (Bradford et al. 1994; Fisher & Shaffer 1996), suggesting that additional factors are involved. As field data on introduced predators and information on the basic biology of pathogens accumulates, spatial analysis can help in the future to evaluate the importance of both disease and exotic species.

Conclusions

Our analysis indicates that multiple factors may be responsible for declines of California amphibians. For the species inhabiting vernal pools (*A. californiense, S. hammondii*), declines were strongly associated with habitat alteration, principally in the form of surrounding urban and, to a lesser extent, agricultural land use. *Bufo californicus* is a unique habitat specialist that also showed a strong association of decline with habitat alteration, although its habitat has been less heavily altered by urbanization and agriculture. For the remaining five species, declines were generally not consistent with those expected from the UV-B or climate-change hypotheses, although there is partial support for each hypothesis in a single species. Declines for four of the five species were strongly associated with the amount of upwind agricultural land use, suggesting that wind-borne agrochemicals may be a factor.

For each of the hypotheses, we have chosen to examine the simplest set of biologically meaningful predictions that we can envision. We believe that it is worth examining these simple predictions before searching for more complex patterns. This strategy has, in some cases, already successfully associated distributional changes with a hypothesized cause. For example, Parmesan (1996) found that declines of a butterfly on the Pacific coast of North America fit the simple climate-change model of range change. Clearly, each of the four factors we examined may affect the pattern of decline in complex ways that might not be detected by our analysis. For example, Pounds et al. (1999) linked declines of montane amphibians at Monte Verde, Costa Rica, to changes in daily precipitation associated with global climate change.

Linking observed patterns to presumed underlying processes must be undertaken with caution because multiple processes can generate similar patterns. And given potential confounding factors, the absence of a pattern should not be taken as proof of the absence of a process. Climate change and UV-B radiation may be con-

tributing to amphibian declines in California, despite our generally negative results for these hypotheses. Furthermore, multiple factors may interact to cause declines. For example, exposure to pesticides may weaken immune systems, increasing susceptibility to disease (Taylor et al. 1999*a*), and the presence or absence of predators can radically change the toxicity of pesticides (Relyea & Mills 2001). Nonetheless, we believe that spatial analysis is a valuable approach for examining large-scale observational data and provides a powerful statistical framework within which to associate declines with plausible mechanisms.

Field and laboratory experiments on individual organisms are vital to understanding possible mechanisms causing declines, but such experiments are necessarily restricted to individuals or small local populations. Population changes above the local site level cannot be subjected to experiments and can be quantified and analyzed only through large-scale observational studies. Under a reasonable set of biological assumptions, the spatial analysis we conducted generated clear predictions that can be tested with field and laboratory studies. Specifically our work should encourage further investigation into the role of agrochemicals in amphibian declines. Pesticides have been suggested as a cause of decline (Carey & Bryant 1995; Stebbins & Cohen 1995; Drost & Fellers 1996; Lips 1998; Green & Kagarise Sherman 2001), and recent experimental research on amphibian species in the United States lends support to this suggestion (Taylor et al. 1999*a*; Boone & Semlitsch 2001; Relyea & Mills 2001). To date there has been little field research on this subject. For example, in both Central America (Pounds & Crump 1994; Lips 1998; Lips 1999) and Australia (Richards et al. 1993), major amphibian declines have occurred in areas close to and downwind, for part of the year, from large agricultural zones. Yet little research on contaminants has been conducted in these areas.

at UCD, the UCD Agricultural Experiment Station, and the National Science Foundation.

Literature Cited

Adams, M. J. 1999. Correlated factors in amphibian decline: Exotic species and habitat change in western Washington. Journal of Wildlife Management **63:**1162-1171.

Alexander, M. A., and J. K. Eischeid. 2001. Climate variability in regions of amphibian declines. Conservation Biology **15:**930-942.

Aston, L. S., and J. N. Seiber. 1997. Fate of summertime airborne organophosphate pesticide residues in the Sierra Nevada mountains. Journal of Environmental Quality **26:**1483-1492.

Berger, L., R. Speare, P. Daszak, D. E. Green, A. A. Cunningham, C. L. Goggin, R. Slocombe, M. A. Ragan, A. D. Hyatt, K. R. McDonald, H. B. Hines, K. R. Lips, G. Marantelli, and H. Parkes. 1998. Chytridiomycosis causes amphibian mortality associated with population declines in the rain forests of Australia and Central America. Proceedings of the National Academy of Sciences of the United States of America **95:**9031-9036.

Blaustein, A. R., D. G. Hokit, R. K. Ohara, and R. A. Holt. 1994. Pathogenic fungus contributes to amphibian losses in the Pacific Northwest. Biological Conservation **67:**251-254.

Blumthaler, M. 1993. Solar UV measurements. Pages 17-79 in M. Tevini, editor. Environmental effects of UV (ultraviolet) radiation. Lewis Publisher, Boca Raton, Florida.

Boone, M. D., and R. D. Semlitsch. 2001. Interactions of an insecticide with larval density and predation in experimental amphibian communities. Conservation Biology **15:**228-238.

Bradford, D. F. 1989. Allotopic distribution of native frogs and introduced fishes in high Sierra Nevada lakes of California: implications of the negative impact of fish introductions. Copeia **1989:**775-778.

Bradford, D. F., D. M. Graber, and F. Tabatabai. 1994. Population declines of the native frog, *Rana muscosa*, in Sequoia and Kings Canyon National Parks, California. Southwestern Naturalist **39:**323-327.

Cabrera, S., S. Bozzo, and H. Fuenzalida. 1995. Variations in UV radiation in Chile. Journal of Photochemistry and Photobiology **28:**137-142.

Carey, C., and C. J. Bryant. 1995. Possible interrelations among environmental toxicants, amphibian development, and decline of amphibian populations. Environmental Health Perspectives **103:**13-17.

Carey, C., N. Cohen, and L. Rollins-Smith. 1999. Amphibian declines: an immunological perspective. Developmental and Comparative Immunology **23:**459-472.

Carey, C., W. R. Heyer, J. Wilkinson, R. A. Alford, J. W. Arntzen, T. Halliday, L. Hungerford, K. R. Lips, E. M. Middleton, S. A. Orchard, and A. S. Rand. 2001. Amphibian declines and environmental change: use of remote-sensing data to identify environmental correlates. Conservation Biology **15:**903-913.

Cory, L., P. Fjerd, and W. Serat. 1970. Distribution patterns of DDT residues in the Sierra Nevada mountains. Pesticide Monitoring Journal **3:**204-211.

Crooks, K. R., and M. E. Soulé. 1999. Mesopredator release and avifaunal extinctions in a fragmented system. Nature **400:**563-566.

Cunningham, A. A., T. E. S. Langton, P. M. Bennett, J. F. Lewin, S. E. N. Drury, R. E. Gough, and S. K. MacGregor. 1996. Pathological and microbiological findings from incidents of unusual mortality of the common frog (*Rana temporaria*). Philosophical Transactions of the Royal Society of London, B **351:**1539-1557.

Datta, S., L. Hansen, L. McConnell, J. Baker, J. LeNoir, and J. N. Seiber. 1998. Pesticides and PCB contaminants in fish and tadpoles from the Kaweah River Basin, California. Bulletin of Environmental Contamination and Toxicology **60:**829-836.

Davidson, C., H. B. Shaffer, and M. R. Jennings. 2001. Declines of the California red-legged frog: climate, UV-B, habitat and pesticides hypotheses. Ecological Applications **11:**464-479.

Department of Pesticide Regulation (DPR). 1990. Pesticide use report: 1990. DPR, Sacramento, California.

Department of Pesticide Regulation (DPR). 1994. Pesticide use report: 1994. DPR, Sacramento, California.

Department of Pesticide Regulation (DPR). 1998. Pesticide use report: 1995. DPR, Sacramento, California.

Drost, C. A., and G. M. Fellers. 1996. Collapse of a regional frog fauna in the Yosemite area of the California Sierra Nevada, USA. Conservation Biology **10:**414-425.

Fahrig, L., J. H. Pedlar, S. E. Pope, P. D. Taylor, and J. F. Wegner. 1995. Effect of road traffic on amphibian density. Biological Conservation **73:**177-182.

Fellers, G. M., and C. A. Drost. 1993. Disappearance of the Cascades frog *Rana cascadae* at the southern end of its range, California, USA. Biological Conservation **65:**177-181.

Fisher, R. N., and H. B. Shaffer. 1996. The decline of amphibians in California's Great Central Valley. Conservation Biology **10:**1387-1397.

Green, D. E., and C. Kagarise Sherman. 2001. Diagnostic histological findings in Yosemite toads (*Bufo canorus*) from a die-off in the 1970s. Journal of Herpetology **35:**92-103.

Gross, S. J., and T. D. Price. 2000. Determinants of the northern and southern range limits of a warbler. Journal of Biogeography **27:** 869-878.

Hayes, M. P., and M. R. Jennings. 1986. Decline of ranid frog species in western North America: are bullfrogs (*Rana catesbeiana*) responsible? Journal of Herpetology **20:**490-509.

Hayes, T. P., J. J. R. Kinney, and N. J. M. Wheeler. 1984. California surface wind climatology. California Air Resources Board, Sacramento.

Herman, J. R., N. Krotkov, E. Celarier, D. Larko, and G. Labow. 1999. Distribution of UV radiation at the Earth's surface from TOMS-measured UV-backscattered radiances. Journal of Geophysical Research **104:**12,059-12,076.

Hosmer, D. W., and S. Lemeshow. 1989. Applied logistic regression. Wiley, New York.

Jennings, M. R. 1988. Natural history and decline of native ranids in California. Pages 61-72 in H. F. De Lisle, P. R. Brown, B. Kaufman, and B. M. McGurty, editors. Proceedings of the conference on California herpetology. Southwestern Herpetologists Society, Van Nuys, California.

Jennings, M. R., and M. P. Hayes. 1994. Amphibian and reptile species of special concern in California. California Department of Fish and Game, Inland Fisheries Division, Rancho Cordova.

Karl, R. T., R. W. Knight, R. R. Easterling, and R. G. Quale. 1996. Indices for climate change for the United States. Bulletin of the American Meteorological Society **77:**279-292.

Kiesecker, J. M., and A. R. Blaustein. 1995. Synergism between UV-B radiation and a pathogen magnifies amphibian embryo mortality in nature. Proceedings of the National Academy of Sciences of the United States of America **92:**11049-11052.

Kiesecker, J. M., A. R. Blaustein, and L. K. Belden. 2001*a*. Complex causes of amphibian population declines. Nature **410:**681-683.

Kiesecker, J. M., A. R. Blaustein, and C. I. Miller. 2001*b*. Transfer of a pathogen from fish to amphibians. Conservation Biology **15:**1064-1070.

Kirkpatrick, M., and N. H. Barton. 1997. Evolution of a species' range. The American Naturalist **150:**1-23.

Knapp, R. A., and K. R. Matthews. 2000. Non-native fish introductions and the decline of the mountain yellow-legged frog from within protected areas. Conservation Biology **14:**428-438.

Kupferberg, S. J. 1997. Bullfrog (*Rana catesbeiana*) invasion of a California river: the role of larval competition. Ecology **78:**1736-1751.

Laurance, W. F. 1996. Catastrophic declines of Australian rainforest frogs: Is unusual weather responsible? Biological Conservation **77:** 203-212.

Lawler, S. P., D. Dritz, T. Strange, and M. Holyoak. 1999. Effects of introduced mosquitofish and bullfrogs on the threatened California red-legged frog. Conservation Biology **13:**613-622.

LeNoir, J. S., L. L. McConnell, M. G. Fellers, T. M. Cahill, and J. N.

Seiber. 1999. Summertime transport of current-use pesticides from California's Central Valley to the Sierra Nevada mountain range, USA. Environmental Toxicology and Chemistry **18:**2715–2722.

Lips, K. R. 1998. Decline of a tropical montane amphibian fauna. Conservation Biology **12:**106–117.

Lips, K. R. 1999. Mass mortality and population declines of anurans at an upland site in western Panama. Conservation Biology **13:**117–125.

Madronich, S., R. L. McKenzie, M. M. Caldwell, and L. O. Bjorn. 1995. Changes in ultraviolet radiation reaching the earth's surface. Ambio **24:**143–152.

Marco, A., C. Quilchano, and A. R. Blaustein. 1999. Sensitivity to nitrate and nitrite in pond-breeding amphibians from the Pacific Northwest, USA. Environmental Toxicology and Chemistry **18:**2836–2839.

Marsh, D. M., and P. B. Pearman. 1997. Effects of habitat fragmentation on the abundance of two species of Leptodactyliid frogs in an Andean montane forest. Conservation Biology **11:**1323–1328.

McConnell, L. L., J. S. LeNoir, S. Datta, and J. N. Seiber. 1998. Wet deposition of current-use pesticides in the Sierra Nevada mountain range, California, USA. Environmental Toxicology and Chemistry **17:**1908–1916.

Middleton, E. M., J. R. Herman, E. A. Celarier, J. Wilkinson, C. Carey, and R. J. Rusin. 2001. Evaluating ultraviolet radiation exposure with satellite data at sites of amphibian declines in Central and South America. Conservation Biology **15:**914–929.

Parmesan, C. 1996. Climate and species' range. Nature **382:**765–766.

Peters, R. L. 1991. Consequences of global warming on biological diversity. Pages 99–118 in R. L. Wyman, editor. Global climate change and life on earth. Chapman Hall, New York.

Pounds, J. A., and M. L. Crump. 1994. Amphibian declines and climate disturbance: the case of the golden toad and the harlequin frog. Conservation Biology **8:**72–85.

Pounds, J. A., M. P. L. Fogden, and J. H. Campbell. 1999. Biological response to climate change on a tropical mountain. Nature **398:**611–614.

Relyea, R. A., and M. Mills. 2001. Predator-induced stress makes the pesticide carbaryl more deadly to gray treefrog tadpoles. Proceedings of the National Academy of Science of the United States of America **98:**2491–2496.

Richards, S. J., K. R. McDonald, and R. A. Alford. 1993. Declines in populations of Australia's endemic tropical rainforest frogs. Pacific Conservation Biology **1:**66–77.

Shaffer, H. B., and P. C. Trenham. 2001. Species account: *Ambystoma californiense*. In M. J. Lannoo, editor. Status and conservation of United States amphibians. University of California Press, Berkeley.

Shaffer, H. B., R. N. Fisher, and C. Davidson. 1998. The role of natural history collections in documenting species declines. Trends in Ecology & Evolution **13:**27–30.

Sokal, R. R., and F. J. Rohlf. 1995. Biometry. W. H. Freeman, New York.

Stebbins, R. C. 1951. Amphibians of western North America. University of California Press, Berkeley.

Stebbins, R. C., and N. W. Cohen. 1995. A natural history of amphibians. Princeton University Press, Princeton, New Jersey.

Taylor, S. K., E. S. Williams, and K. W. Mills. 1999*a*. Effects of malathion on disease susceptibility in Woodhouse's toads. Journal of Wildlife Diseases **35:**536–541.

Taylor, S. K., E. S. Williams, E. T. Thorne, K. W. Mills, D. I. Withers, and A. C. Pier. 1999*b*. Causes of mortality of the Wyoming toad. Journal of Wildlife Diseases **35:**49–57.

Trenham, P. C., H. B. Shaffer, W. D. Koenig, and M. R. Stromberg. 2000. Life history and demographic variation in the California tiger salamander (*Ambystoma californiense*). Copeia **2000:**365–377.

Vos, C. C., and J. P. Chardon. 1998. Effects of habitat fragmentation and road density on the distribution pattern of the moor frog *Rana arvalis*. Journal of Applied Ecology **35:**44–56.

Wake, D. B. 1991. Declining amphibian populations. Science **253:**860.

Zabik, J. M., and J. N. Seiber. 1993. Atmospheric transport of organophosphate pesticides from California's Central Valley to the Sierra Nevada Mountains. Journal of Environmental Quality **22:**80–90.