

A close-up photograph of a green frog with black spots perched on a tree branch. The frog is the central focus, with its body and limbs clearly visible. The background is a soft, out-of-focus green, suggesting a natural habitat. The text is overlaid on the image in a bold, yellow font.

AMPHIBIANS IN DECLINE
CANADIAN STUDIES OF A GLOBAL PROBLEM

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ANURAN SPECIES RICHNESS IN AGRICULTURAL LANDSCAPES OF QUÉBEC: FORESEEING LONG- TERM RESULTS OF ROAD CALL SURVEYS

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ABSTRACT.—Breeding call surveys were conducted at 157 sites in southern Québec to assess the relationship between agricultural landscape features and the occurrence of 9 species of anurans. Using canonical discriminant analyses, the presence of 5 species—*Pseudacris crucifer*, *Rana sylvatica*, *Rana pipiens*, *Hyla versicolor*, and *Rana clamitans*—was related to ≥ 1 terrestrial or aquatic landscape features. The toad, *Bufo americanus*, was ubiquitous while 3 other species were too scarce to draw any conclusions about their preferences. In general, species richness was inversely related to the occurrence of monocultures where pesticides were usually applied. The effect of increased monoculture, however, could not be dissociated from the loss of essential habitats such as permanent waterbodies, forests, and old fields. These landscape features have diminished during the past 25 yr and it is highly probable that anuran populations have changed correspondingly. In our results, > 65% of the variability in anuran occurrences remained unexplained. This might be partly due to the crude nature of the data obtained from the road call surveys and the landscape features evaluations. To investigate factors involved in a global decline of amphibians, we will need more detailed data on habitat and breeding locations, together with sampling strategies oriented toward testing hypotheses.

RÉSUMÉ.—Des inventaires de chants de reproduction ont été réalisés dans le sud du Québec pour déterminer la relation entre les éléments du paysage agricole et la présence de 9 espèces d'anoures. À partir d'analyses canoniques discriminantes, la présence de 5 espèces, *Pseudacris crucifer*, *Rana sylvatica*, *Rana pipiens*, *Hyla versicolor*, et *Rana clamitans*, fut associée à un ou plusieurs éléments terrestres ou aquatiques. Le crapaud, *Bufo americanus*, était ubiquiste tandis que 3 autres espèces n'étaient pas assez fréquentes pour tirer des conclusions sur leurs préférences. En général, la richesse spécifique décroissait avec l'augmentation de la superficie en monoculture où des pesticides sont généralement appliqués. Toutefois, l'effet d'une augmentation de la monoculture ne pouvait être dissocié de la perte d'habitats essentiels (plan d'eau permanent, forêt et friche). Des changements dans ces éléments du paysage sont survenus au cours des 25 dernières années et il est fort probable que les populations d'anoures aient également changé. Dans nos résultats, une grande part de la variabilité dans la présence des anoures est demeurée inexplicée (> 65%). Cela pourrait être dû en partie à l'imprécision des données provenant de l'inventaire routier des chants de reproduction et de l'évaluation des éléments du paysage. Afin de rechercher les facteurs impliqués dans un déclin global des amphibiens, nous aurons besoin de données plus détaillées sur les habitats et les sites de reproduction, ainsi que des stratégies d'échantillonnage permettant la vérification d'hypothèses.

In the past 50 yr, the intensification of agriculture in southern Québec has modified the landscape and adversely affected wildlife (Freemark and Boutin 1994; Boutin et al. 1994; DesGranges et al.

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1995). In the most productive lands of the Saint Lawrence Lowlands, wetlands and woodlands have been replaced by large scale agricultural monocultures. Irrigation has resulted in the replacement of natural wetland habitats by temporary ditches. Corn production, which was favoured by subsidies for underground drainage, increased in Québec from 43,000 ha in 1973 to 228,000 ha in 1987 (Jobin et al. 1994). On the other hand, cultivation of marginal lands located in the foothills of the Appalachians and the Laurentians has been partly abandoned, leaving old fields and forests in their place (Boutin et al. 1994; Jobin et al. 1994).

Ten species of anurans live in southern Québec (Bider and Matte 1994). They require permanent or temporary waterbodies for breeding (Collins and Wilbur 1979), and marshes, fields or forests for summer foraging. Anurans may be unable to find suitable habitats in the present agricultural landscape. This has been the case in the Netherlands where a decline in amphibians was noticed after a decrease in the number of pools, mainly drinking ponds for cattle, between 1960 and 1980. Now, out of 12 native species, 2 are extinct and 5 are endangered (Laan and Verboom 1990). Berger (1989) noted a similar decline of amphibians in the agricultural landscape of Poland, and found that chemical pollution of aquatic habitats caused the death of larvae.

The recent awareness of a possible amphibian declines in Canada raised the need for population monitoring (Bishop and Pettit 1992). Programs based on breeding call surveys were recently set up in various part of the country. Though methodological and analytic aspects have been addressed (Bishop et al., this volume; Lepage et al., this volume), little is known on the usefulness of this approach, especially to identify causes of decline.

The objectives of this study were to determine anuran species richness in agricultural lands, and to infer the conditions required for their survival. For those purposes, we applied the methodology of breeding call surveys over an area where conditions varied from rustic and heterogeneous landscape to large scale monocultures. We felt that this range of habitat conditions would reflect the extent of landscape changes that occurred in southern Québec since agricultural use intensified. Therefore, we expected to find indications of a possible decline of anuran in the past, as well as information concerning the usefulness of road call surveys to evaluate causes of decline.



Bullfrog, *Rana catesbeiana*. Photo by John Mitchell

MATERIAL AND METHODS

We conducted the survey along 10 Breeding Bird Survey (BBS) routes used to study the response of birds to changes in agricultural landscapes over the past 25 yr (Jobin et al. 1994). Most routes were located in the productive St. Lawrence lowlands, but some routes reached marginal lands located in the foothills of the Appalachians and the Laurentians (Fig. 1). Most routes were within the range of 10 species of anurans (Bider and Matte 1994). From each BBS route, consisting of 50 stops set at every 800 m, we selected some stops according to the landscape features present, as described in Jobin et al. (1994).

Following the methodology described in Lepage et al. (this volume), we conducted anuran call counts after sunset, during warm nights. We listened for 3 min at each stop, noting the abundance of each species that was calling from any direction. We ranked abundance by using a calling index: 0 = no calls, 1 = few males calling, 2 = calls overlapping but some individual calls still distinguished from the chorus, 3 = a full chorus. We repeated surveys during the year to correspond with the breed-

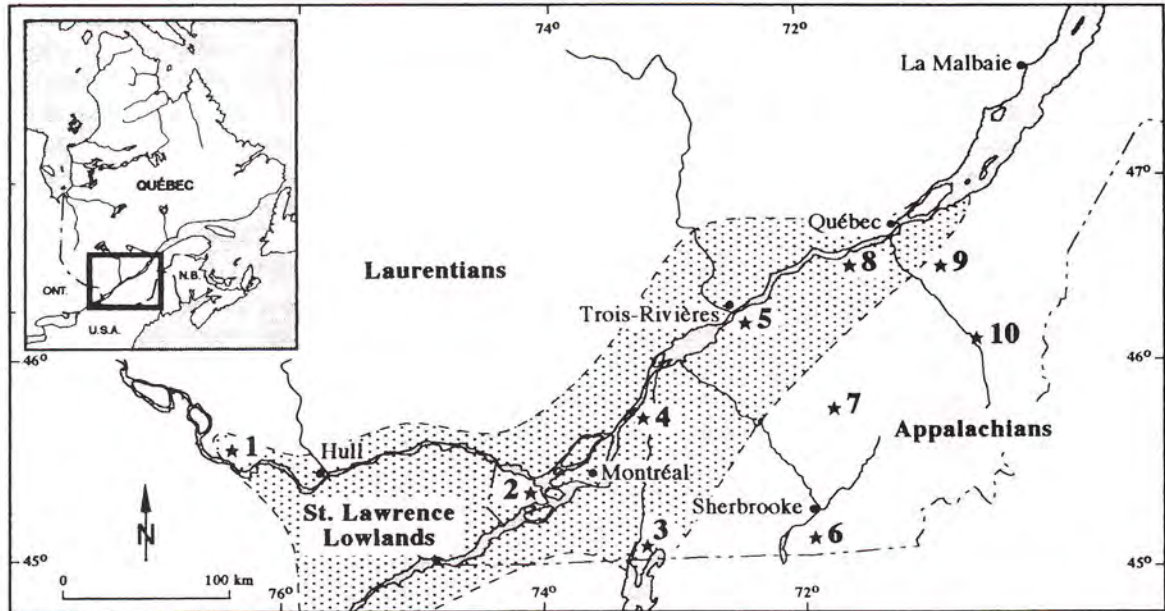


Figure 1. Location of routes surveyed in 1992 and 1993 in southern Québec.

ing period of each species (Lepage et al., this volume). If there was too much wind, or if the temperature dropped suddenly and the frogs stopped calling, we re-surveyed the route the next suitable night. We neglected stops where other noises from cars or barn fans impaired our ability to hear. In 1992, 157 stops were surveyed (Table 1), and we re-surveyed most routes at least once in 1993 to detect either the early breeding species like *Rana sylvatica* or other species that had been missed the 1st time. We surveyed routes 1, 3, and 4 more frequently in 1993 to measure variation in the presence and abundance of species from one year to another.

The presence of each species was related to landscape features by the mean of canonical discriminant analyses. Variables included the presence or absence of permanent waterbodies or slow flowing watercourses (slope < 10 m over 500 m), intermittent slow flowing watercourses, old fields, and forests identified using recent aerial photographs (1:15,000) and topographical maps (1:20,000). These features were counted if within 400 m from the listening stop. Forests located up to 100 m away from the 400 m radius were also taken into account on the assumption that frogs could return to these forests after breeding. The routes which were located out of the distribution range of a species or which were not surveyed during the species breeding period, were removed from the analysis for that species.

Species occurrence and richness (total number of species) were also compared to the percentage of the area in monoculture and the size of wood lots. We identified crops in the field during 1992 (corn, wheat, oat, barley, soybean, or vegetables) and we evaluated the percentage of

Table 1. Number of stops and dates of surveys for each route in 1992 and 1993.

Route	Stops	Survey Dates	
		1992	1993
1. Eardley	13	8 May; 2 June	29 April; 8,14,30 May; 6 July
2. Hudson	6	7 May, 14 June; 10 July	
3. Lacolle	16	29 April; 10, 30 May; 3 July	25 April; 6 May; 2,7,13 July
4. Verchères	25	28 April; 9 May; 1,30 June	28 April; 10 May; 11 June; 7 July
5. St-Célestin	22	13 May; 30 May	30 April
6. Coaticook	13	20 May; 17 June	2 May
7. Ham Sud	10	20 May; 17 June	2 May
8. St-Antoine	29	12 May; 8 June	26 May; 29 June
9. Ste-Marguerite	18	11 May; 18 June	
10. Morisset	5	11 May; 18 June	

the area of each stop represented by monocultures in which pesticides were usually applied. We distinguished stops with small wood lots (< 5 ha, $n = 25$) from those with woodlands of greater area ($n = 82$). Fifty stops had no forest at all. We used contingency table analyses (Fisher's exact test) to evaluate the relationship between a landscape variable and the species presence. Rank tests were used when the variables were not binary (percent of area in monocultures, and species richness).

We calculated rank-correlations (Kendall's τ corrected for ties [Kendall and Gibbons 1990]) among habitat variables in order to determine how these variables were linked together and if multicollinearity (Pedhazur 1982) could have impaired their simultaneous analysis by the canonical discriminant method. The test of significance corresponded to the Mann-Whitney U test when one of the correlated variables was dichotomous, while it was equivalent to Fisher's exact test when both variables were dichotomous (Burr 1960).

RESULTS

Nine species were heard during the 2 yr of survey. *Pseudacris crucifer* was found at 135 stops (86% of all stops), *Bufo americanus* at 133 (85%), *R. sylvatica* at 80 (51%), *Rana clamitans* at 56 (36%), *Rana pipiens* at 41 (26%), *Hyla versicolor* at 28 (18%, or 34% of the locations within its distribution range), *Rana catesbeiana* at 8 (5%), *Rana septentrionalis* at 4 (3%), and *Pseudacris triseriata* at 2 (1%, or 3 to 6% of the locations within its distribution range).

Calling index values varied greatly between years. Different values were obtained for 49 to 60% of the stops surveyed both years for *P. crucifer* (51%, $n = 41$ stops where the species was present ≥ 1 yr), *B. americanus* (49%, $n = 30$), and *R. pipiens* (60%, $n = 55$). The estimated abundance at 1 stop varied from one yr to another but no general trends were noticed. On the other hand, presence/absence data differed in 11 to 50% of the stops: 22% for *P. crucifer*, 11% for *B. americanus*, and 50% for *R. pipiens*. This suggests that presence/absence would be a more stable index than that of abundance and we therefore used only presence/absence data in the subsequent analysis.

In intensive agriculture, wildlife habitats (forest, old field, wetland) were rarer while intermittent ditches were more frequent (Fig. 2). The correlation of the percent of land in monoculture with the presence of all other variables precluded any conclusion about its direct effect on frog occurrence. We had to remove the variable "percent of monoculture" from the canonical discriminant analysis in

order to limit the problem of multicollinearity (e.g. Pedhazur 1982). Therefore, each variable used in the analysis represented a complex of variables that subsumed the direct effects of monoculture on anurans.

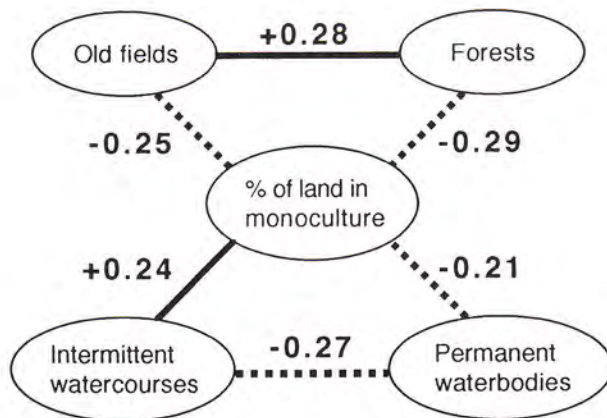


Figure 2. Significant correlations (Kendall τ , $P < 0.001$) among landscape features of the 157 stops surveyed in agricultural lands of southern Québec.

Contingency table analyses and canonical discriminant analyses (CDA) gave concordant results about the relationship between species and landscapes (Table 2). *Bufo americanus*, was ubiquitous and its presence was unrelated to any landscape feature (CDA and Fisher's exact tests, $P < 0.05$). The occurrence of 5 frog species was significantly related to ≥ 1 landscape features (CDA, $P < 0.001$), though a large portion of the variability in species presence or absence remained unexplained ($R^2 < 0.35$ in all cases). The presence of *R. sylvatica* was related solely to the presence of forest, while *H. versicolor* presence was related to forest and, to a lesser extent, old field (Table

Table 2. Relative importance of landscape features (standardized coefficients) in the canonical variates discriminating for the presence or absence of the 6 species considered. *R* is the canonical correlation coefficient.

Species	Stations	Landscape Features				R	P
		Intermittent watercourse	Permanent waterbody	Old field	Forest		
<i>Pseudacris crucifer</i>	157	-0.22	-0.17	0.61*	0.55*	0.43	< 0.001
<i>Rana sylvatica</i>	151	0.19	0.24	-0.08	0.98*	0.42	< 0.001
<i>Rana pipiens</i>	134	0.58*	0.62*	0.40	0.41	0.42	< 0.001
<i>Hyla versicolor</i>	82	-0.22	0.02	0.38*	0.81*	0.58	< 0.001
<i>Rana clamitans</i>	157	0.34	0.60*	0.43*	0.57*	0.41	< 0.001
<i>Bufo americanus</i>	157	-0.82	0.55	0.49	0.22	0.07	> 0.05

*Significant relationship ($P < 0.05$) between the presence of a species and the occurrence of a landscape feature

2). *Pseudacris crucifer* presence was related to old field and, secondarily, to forest. The presence of *R. clamitans* was related to the presence of permanent waterbodies and, as well, to the presence of forest and old field. *Rana pipiens* was associated mainly with permanent and intermittent waterbodies. No statistical analysis could be performed for the 3 less common species because of small sample sizes. *Rana catesbeiana* and *R. septentrionalis* were encountered in a few permanent waterbodies while *P. triseriata* was found in a pasture and a hay field located in marginal lands.

Pseudacris crucifer, *R. sylvatica*, and *R. clamitans* were usually found where a low percentage of the area was in monoculture (Mann-Whitney U , $P < 0.05$ in all tests). Species richness at each stop was also negatively correlated to the percentage of the area in monoculture (Kendall τ , $P < 0.001$). The number of species tended to be higher where there were permanent waterbodies, old fields, and forests (Mann-Whitney U , $P < 0.01$ in all tests). None of the species was found less frequently at stops adjoining only small woodlots < 5 ha than at stops with larger forests (Fisher's exact test, $P < 0.05$ in all tests).

DISCUSSION

The strong relationships between landscape features and the presence of many anuran species indicate that forest and aquatic habitats are prerequisites for survival. These predictable relationships represent known habitat preferences of each species (e.g. Collins and Wilbur 1979). Beebee (1985) also found similar facts using discriminant analysis to determine habitat preference of amphibians in south-east England.

Because correlations among landscape features (Fig. 2) agreed with the history of agricultural changes, the geographical gradient surveyed reflected the temporal trends in agricultural practices during the past 25 yr, as described by Jobin et al. (1994). We therefore suspect that frog occurrences also varied over this period. In the lowlands, intensification of agriculture has probably affected frog populations by the loss of habitat. Although fewer species were usually found at stops with large scale monocultures, the direct effect of monocultures and associated pesticides could not be evaluated because of the strong correlations with all other landscape features measured. Nevertheless, the association between the semi-riparian species, *R. clamitans* (Martof 1953) and the presence of forest and old field might reflect that waterbodies in rustic landscapes were more suitable than those in areas of intensive agriculture. The wide use of herbicides to maintain agricultural monocultures may degrade habitat quality for anurans by reducing the diversity of plants, soil organisms, and other invertebrates (Sotherton et al. 1988). In addition, chemical pollution of aquatic habitat may affect survival of anuran larvae (Berger 1989). Genetical damages and diseases were also found in *R. clamitans* populations from cultivated lands of southern Québec (Bonin et al., this volume).

The level of forest fragmentation (decreased to wood lots ≤ 5 ha), did not seem to affect the occurrence of woodland species of frogs. We selected 5 ha to have a sufficient number of stops with small wood lots, but this might not be a critical threshold. Both forest history and their distribution in agricultural landscapes can influence amphibian presence (Bonin 1991; Waldick, this volume) and dispersal (Laan and Verboom 1990) and should be considered in studying the impact of forest fragmentation on amphibians. Over the last 25 yr in Québec, forest loss and fragmentation took place in the most productive agricultural lands (Jobin et al. 1994). In the marginal lands, the increase of forested areas may benefit woodland frogs, but could not be verified in this study since forests were already present 25 yr ago in most of the marginal lands surveyed.

Usefulness Of Road Call Surveys For Detecting Causes Of Decline

Although the road call surveys permit large areas to be surveyed and geographical variations in species distribution and abundance to be noted (Lepage et al., this volume), there are many limitations in using this method to assess causes of decline. With the road call count technique used, it was rarely possible to locate the chorus from the listening point and, therefore, breeding habitats remained unknown. Habitat characteristics had to be restricted to broad landscape features discernible on aerial photographs or on maps. Therefore, temporary woodland ponds and streams had to be neglected. Intermittent watercourses were distinguished from permanent ones following indications on maps. In some instances, calls could have been heard from a breeding site located out of the 400-m radius and toads could have been heard calling while migrating through the stop toward a breeding site located outside of the survey area. Inter-year variations in calling data were another source of error with this method. Variation in climate was probably the major factor affecting road call count efficiency to evaluate amphibian presence or abundance. For example, 1992 was a bad year to survey *H. versicolor* in a breeding population in southern Ontario (Bertram and Berrill, this volume). Full choruses were seldom heard because of cool temperatures during the species usual breeding period. In 1992, a few males called during several good nights spread over 8 wk, while in 1993, a larger chorus could be heard on many nights within a shorter breeding period (Bertram and Berrill, this volume). From our results, the use of presence/absence data rather than estimated abundance with the calling index reduced the inter-year variation and, therefore, was considered more accurate.

The efficiency of the call counting technique varied also among locations and among species. Listening conditions vary from one site to another because of topography, buildings, wood lots, or simply the presence of a nearby chorus which masks the sound of frogs calling from further away. Changes in listening conditions at the stops will probably result in erroneous fluctuations in calling data. Species having loud calls, and breeding over a long period, have more chance to be detected, as is the case for the 2 most common species, *P. crucifer* and *B. americanus*. This bias may be increased by the distance at which calls are required to be heard (up to 400 m).

Our study using road call surveys pointed out the obvious influence of habitat loss on anuran survival. However, only a low percentage ($R^2 = 0.17$ to 0.34) of the variability associated with frog presence was explained by the landscape features used in the canonical discriminant analysis. This might be due to the crude nature of the data collected, the omission of important factors such as habitat characteristics, pollutants, zoogeography or interspecific relations, or to the ecological plasticity of each species. Hecnar (this volume), who built predictive models of amphibian diversity in farmland ponds of Southern Ontario, was also left with much unexplained variability (about 60%), even though he considered numerous habitat variables from breeding sites and surrounding areas. Similarly, Beebee (1985) found that landscape features were better predictors of amphibian presence than most measurements relating specifically to breeding ponds. Beebee (1985) and Hecnar (this volume) had canonical correlation levels comparable to ours even if they established amphibian presence from captures at breeding ponds rather than from road call surveys. This suggests the variability depends on other factors than low precision in habitat and population assessment inherent to road call surveys.

Ever since the recent reports of unexplained die-offs and decline in various amphibian populations throughout the world (Wake 1991), scientists and wildlife managers have been seeking to answer if

there is a global trend not simply due to the loss of habitat. The road call surveys initiated in Canada (Bishop et al., this volume, Lepage et al., this volume) might not contribute to that question mainly because they will be conducted in areas where changes in habitats and landscapes are highly probable in the future. These habitat changes will bring fluctuations in anuran populations and hence might mask the effect of other global factors. To investigate the presumed roles of pesticides, atmospheric pollutants, UV-B radiation etc. in a global decline of amphibians, we need good data on these factors and on breeding habitat conditions. Furthermore, a sampling strategy would have to be oriented toward testing hypotheses in order to investigate matters such as the effect of pesticides on species occurrence. A long-term monitoring program based on the road call count technique without a sampling strategy would not provide any more valuable information on the causes of decline than the few predictable results presented here for agricultural lands.

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American toad, Bufo americanus. Photo by Donald F. McAlpine.