UNIVERSITÉ DU QUÉBEC

MÉMOIRE DE RECHERCHE PRÉSENTÉ À
L'UNIVERSITÉ DU QUÉBEC À TROIS-RIVIÈRES

COMME EXIGENCE PARTIELLE
DE LA MAÎTRISE EN SCIENCES DE L'ENVIRONNEMENT

PAR

DANIEL POULIOT

DÉVELOPPEMENT, CROISSANCE ET ENVIRONNEMENT DES TÊTARDS DE GRENOUILLE LÉOPARD DU NORD (*RANA PIPIENS*) DANS LA PLAINE INONDABLE DU LAC SAINT-PIERRE

AOÛT 2007

DÉDICACE

Je dédie mon mémoire
À la douce mémoire de ma mère
Louise Guillemette Pouliot †

REMERCIEMENTS

Une maîtrise est un long chemin sur lequel on apprend beaucoup. Parmi toutes les choses que j'ai apprises, l'une me tient à cœur et mérite d'être soulignée. J'ai appris que, malgré l'exubérance de nos talents, nos actions individuelles sont limitées; que notre potentiel atteint son paroxysme lorsque le travail se fait en équipe, enrichi de collaboration, de discussions et d'amitié. C'est dans cet esprit de profonde reconnaissance que je rédige mon mémoire.

Je tiens d'abord à remercier mon directeur, Jean-Jacques Frenette. Il m'a ouvert la porte de son laboratoire et m'a donné l'opportunité de travailler en parallèle de son grand axe de recherche. Sa confiance et son amitié, couplées aux conseils judicieux de sa direction m'ont permis de compléter avec plaisir ce projet de recherche. Je remercie également toute l'équipe du laboratoire d'écologie des eaux douces, particulièrement (par ordre alphabétique) : Guillaume Blanchette-Martineau, Dominique Deshaies, Jean-François Lapierre, Marianne Lefebvre et Patrice Thibeault pour leur aide inestimable durant les travaux de terrain 2004 et 2005.

Je remercie Jean-Claude Bourgeois et Jean-François Desroches, membres généreux de mon comité de révision. Leurs conseils et appuis ont été très précieux durant toutes ces années.

Merci aussi à Frederick W. Schueler, qui, malgré son emploi du temps fort chargé, a accepté de réviser mon mémoire. Son intimité avec le sujet couplé à sa compréhension de la problématique ont beaucoup enrichi mon travail.

Je remercie chaleureusement ma famille. Particulièrement ma conjointe Nathalie, précieuse conseillère et infatigable oreille. Dans mes élans de création, elle sait à la fois m'encourager et me garder les pieds sur terre. Je souligne l'arrivée toute récente de notre petit Gabriel, soleil quotidien et expression intime de la vie sur terre. Je remercie mes parents, Louise et Ghislain, qui m'ont encouragé dans ma passion et soutenu depuis maintenant plusieurs années. Mes beaux-parents, Nicole G. et Fernand Côté, qui eux aussi m'ont soutenu durant ma maîtrise, notamment dans la construction de mes enclos flottants.

Depuis maintenant 14 ans, j'explore avec passion le monde naturel en compagnie d'amis passionnés et chaleureux, j'en profite pour les remercier (par ordre chronologique) : Ian-Érik Gosselin, Alexandre Bélanger, Jean-Marc Touzet, Grégory Bulté, Raphaël Demers, Marc Bois, Jean-François Desroches, Josiane Bergeron, Martin Savard, Jean-Marc Vallières et Maxime Léveillé. Chacun à sa manière, ils influencent et font évoluer ma passion.

AVANT-PROPOS

Conformément aux articles 136, 137 et 138 du règlement des études des cycles supérieurs, il est possible de présenter les résultats obtenus dans le cadre du programme de 2e cycle en Sciences de l'environnement sous forme d'articles scientifiques.

Il a donc été convenu avec mon directeur, le Dr Jean-Jacques Frenette, que trois articles issus de mon projet de maîtrise seront soumis à des périodiques scientifiques. Ces articles ont été rédigés en anglais pour répondre aux exigences officieuses du monde scientifique. Afin de rendre plus aisé la consultation de ce mémoire par des francophones et ainsi favoriser l'échange de connaissances, un résumé en français est présenté dans ce mémoire, en prélude aux articles plus détaillés.

RÉSUMÉ

Des milieux humides artificiels ont été aménagés dans la plaine d'inondation du lac Saint-Pierre pour fournir à la sauvagine des aires de repos et de nidification. Étant donné leur conception et les pratiques de gestion qui y sont appliquées, ces aménagements sont susceptibles d'offrir à la faune des conditions environnementales différentes de ce que l'on retrouve dans les baies naturelles du lac. Nous avons voulu vérifier le potentiel des aménagements de type « bassin permanent » de répondre aux besoins de la grenouille léopard du Nord (*Rana pipiens*) en fournissant aux têtards un site propice au développement et à la croissance. Nous voulions également mettre en relation l'évolution des conditions environnementales et la croissance des têtards. Pour ce faire, nous avons caractérisé hebdomadairement l'environnement, le développement et la croissance des têtards dans un site aménagé et dans une baie du lac. Nous avons également élevé des têtards provenant des deux origines, dans des enclos flottants placés dans le site aménagé. Les aménagements de type « bassin permanent » permettent aux têtards de compléter leur développement sans anomalie particulière et même de croître davantage que ceux des sites naturels. Ils représentent donc un outil de conservation fort intéressant. La qualité, la quantité de nourriture disponible, ainsi que la densité de têtards expliqueraient la différence dans la croissance. Le régime hydrique pourrait vraisemblablement être considéré comme la cause proximale de ces différences.

TABLE DES MATIÈRES

CHAPITRE 1

CHAPITRE 2

CHAPITRE 3

CHAPITRE 4

LISTE DES TABLEAUX

CHAPITRE 2 : ARTICLE 1

CHAPITRE 3 : ARTICLE 2

LISTE DES FIGURES

CHAPITRE 2 : ARTICLE 1

CHAPITRE 3 : ARTICLE 2

CHAPTRIE 4 : ARTICLE 3

LISTE DES SYMBOLES ET DES ABRÉVIATIONS

PNAGS : Plan nord américain de gestion de la sauvagine

NAWMP: North American waterfowl management plan

FAPAQ : Société de la faune et des parcs du Québec

D.U.C.: Ducks Unlimited Canada

RCCAR : Réseau canadien de conservation des amphibiens et des reptiles

LO(W)ESS : Une courbe l*o(w)ess* est une moyenne courante (running mean), calculée et tracée à partir de l'ensemble des données présentes dans une fenêtre donnée. Elle permet d'explorer graphiquement et de modèliser la relation entre deux variables. Il s'agît d'une méthode robuste face à l'influence des points extrêmes.

SMOOTHING PARAMETER : Le "smoothing parameter" dicte le comportement de la courbe *lo(w)ess* en incluant un nombre plus ou moins élevé d'unités dans chaque fenêtre. Un paramètre de 1.0 produira une courbe très lissée, peu fidèle à la relation entre les variables X et Y. Un paramètre de 0.0 produira une courbe stochastique, fidèle à la relation entre les variables, mais dont il sera difficile de dégager une tendance générale.

TN: Azote total (total nitrogen)

TP: Phosphore total (total phosphorus)

K_d PAR : Coefficient d'extinction de la lumière visible

Chl *a* : Chlorophylle *a*

POM : Matière organique particulaire (Particulate organic matter)

SVL : Longueur museau-cloaque (Snout-vent length)

OFT : Optimal foraging theory

CHAPITRE 1

RÉSUMÉ DU PROJET DE RECHERCHE

INTRODUCTION

Le développement et la croissance des formes larvaires chez les amphibiens sont des sujets complexes, aux multiples facettes. La littérature abondante, publiée depuis des décennies dans des journaux scientifiques diversifiés, en témoigne clairement. La littérature existante traite tant d'anatomie et d'ontogénie, que d'écologie et de physiologie. De nombreux chercheurs ont isolé l'effet d'une ou de quelques variables sur le développement, la croissance, la longueur de la période larvaire et la taille à la métamorphose des larves d'urodèles et des têtards d'anoures. Parmi les variables qui ont un effet sur ces traits d'histoire de vie, les plus étudiées sont : la température de l'eau (Herreid II et Kinney 1967; Harkey et Semlitsch 1988), le régime hydrique (Denver *et al.* 1988), la disponibilité (Steinwascher 1978; Murray 1990; Johnson 1991) et la qualité de la nourriture (Johnson 1991; Kupferberg 1997), le pH (Andrén *et al.* 1988; Cummins 1989), la saturation en oxygène (Bradford et Seymour 1988; Crowder *et al.* 1998) et la densité des larves (Gromko *et al.* 1973; Wilbur 1977; Smith 1983; Berven et Chadra 1988; Murray 1990).

Beaucoup de ces études se sont déroulées en laboratoire, afin d'isoler l'effet d'une variable (approche expérimentale), alors que quelques-unes ont intégré l'ensemble de l'environnement, afin d'obtenir un portrait plus global de l'effet de celui-ci sur les larves/têtards. Nous avons choisi d'utiliser, dans le cadre de ce projet de maîtrise, l'approche environnementale pour étudier le développement et la croissance chez les têtards de grenouille léopard du Nord (*Rana pipiens*).

Au Québec, la grenouille léopard du Nord est toujours considérée comme commune en 2007. Elle présente une vaste répartition, qui s'étend depuis le sud du Québec jusqu'à la Baie James et le Labrador (Desroches et Rodrigue 2004). Elle n'est cependant véritablement abondante qu'à la décharge du lac Champlain ainsi que dans la plaine d'inondation du lac Saint-Pierre (Gilbert *et al.* 1998). L'espèce fait d'ailleurs l'objet d'une chasse importante au lac Saint-Pierre (Daigle et Jutras 2001).

L'abondance de cette espèce dans la région du lac Saint-Pierre n'est pas étrangère à la vaste plaine d'inondation et aux nombreux habitats de reproduction qui lui sont disponibles. La plaine d'inondation représente 18 000 hectares de terre inondée chaque année (Langlois *et al.* 1992). Au printemps, le niveau d'eau augmente et le fleuve inonde une grande diversité de communautés végétales, qui sont depuis la berge vers le large : le marécage arborescent, le marécage arbustif et la prairie humide, le marais peu profond et le marais profond. Graduellement, au cours du printemps et de l'été, le fleuve se retire de ces groupements floristiques.

Bien qu'elle représente encore aujourd'hui la plus grande plaine inondable du fleuve Saint-Laurent, celle-ci a été amputée d'une surface considérable de milieux humides au cours du dernier siècle, notamment suite à des pratiques de drainage ou d'endiguement (Benoît *et al.* 1987).

La perte d'habitats pour la faune et le déclin des effectifs d'espèces gibiers comme la sauvagine, a conduit le gouvernement du Québec et de nombreux partenaires québécois à aménager des milieux humides dans la plaine d'inondation du lac Saint-Pierre (FAPAQ 2003). Ces aménagements ont été réalisés dans le cadre de l'application du plan nord-américain de gestion de la sauvagine (PNAGS).

Ce plan signé par le Canada et les États-Unis en 1986 vise à rétablir les populations de canards et d'oies à leur niveau des années 70 (NAWMP Plan Committee 2004). Les milieux humides aménagés représentent quelques 2490 ha dans la plaine inondable du lac Saint-Pierre (FAPAQ 2003). Différentes stratégies d'aménagements ont été utilisées pour répondre aux besoins de la sauvagine, du rat musqué et de quelques espèces de poissons (D.U.C. et N.O.V.E. 1990; FAPAQ 2003). L'un des aspects qui distingue les différentes conceptions d'aménagements concerne la gestion du niveau d'eau (FAPAQ 2003). Certains aménagements sont en phase avec le fleuve Saint-Laurent, alors que d'en d'autres, le niveau d'eau est géré artificiellement tout au cours de la saison.

Dans ces derniers, deux scénarios : le premier consiste à laisser le milieu en phase avec le Saint-Laurent au printemps, puis à fermer les vannes pour maintenir le niveau d'eau durant une période prolongée. Les vannes sont graduellement ouvertes durant l'été pour finalement faire une vidange complète. Le second scénario consiste à maintenir le niveau d'eau élevé dans de grands milieux humides endigués. L'eau de la fonte de la neige et des précipitations s'y accumule. Durant l'été, par évaporation et transpiration, le niveau d'eau diminue tranquillement. Au besoin, une pompe permet de puiser l'eau du Saint-Laurent et de l'ajouter dans le milieu pour ainsi maintenir le niveau d'eau. Ces derniers aménagements « permanents » sont conçus pour permettre à la sauvagine d'élever ses couvées, et c'est de ce type d'aménagement dont il sera question dans le cadre de ce projet de recherche.

Couplés à la diversité des milieux humides naturels présents, les divers milieux humides aménagés ont transformé la plaine inondable du lac Saint-Pierre en une grande mosaïque d'habitats où la grenouille léopard évolue.

Schueler et Karstad (2000), lors d'une conférence présentée au 5^{e} congrès annuel du Réseau canadien de conservation des amphibiens et des reptiles (RCCAR), soulignaient l'abondance de la grenouille léopard du Nord à proximité des aménagements du plan nord américain de gestion de la sauvagine dans le nord-est de l'Ontario. Ils proposaient que ces milieux artificiels puissent jouer le rôle « d'habitats sources » et pouvaient donc contribuer à la conservation locale de cette espèce.

Puisque l'on sait que la nature des conditions environnementales influence le développement et la croissance des têtards, et que ces mêmes conditions ont un effet sur la taille à la métamorphose et la survie des individus, nous nous sommes demandés : a) si les milieux aménagés de type « permanent » permettaient aux têtards d'y compléter leur développement sans anomalies particulières, b) quelles étaient les similitudes et les différences dans le développement et la croissance des têtards dans un site naturel et un site aménagement de type « permanent », c) quelles étaient les similitudes et les différences des conditions environnementales rencontrées par les têtards.

Finalement, nous nous sommes intéressés lors de la deuxième saison de terrain : d) à la croissance des têtards provenant des deux origines, naturelle et aménagée, lorsqu'élevés en conditions semi-contrôlées dans le même milieu humide aménagé.

Ces interrogations avaient pour objectif d'une part, de vérifier l'hypothèse de Schueler et Karstad, et ainsi d'évaluer le potentiel des aménagements dans la conservation de la grenouille léopard du Nord et, d'autre part, de réaliser une étude fondamentale sur la relation entre les conditions environnementales, le développement et la croissance de cette espèce toujours abondante sur le territoire québécois.

AIRE D'ÉTUDE

Les travaux de terrain se sont déroulés sur la rive sud du lac Saint-Pierre, plus particulièrement dans un site naturel situé à Notre-Dame-de-Pierreville (46°07'N 72°52'O) et dans l'aménagement de la Commune de Baie-du-Febvre, situé à Baie-du-Febvre (46°08'N 72°47'O) (FIGURE 2.1). Le site naturel présente toutes les caractéristiques des berges du lac, c'est-à-dire qu'il présente un gradient de communautés végétales inondées au printemps, qui s'assèchent graduellement au cours de l'été (FIGURE 2.2). Le site aménagé est un bassin permanent construit en 1988 qui présente lui aussi une diversité de communautés végétales (FIGURE 2.3). Le niveau d'eau diminue par évapotranspiration au cours de l'été, mais les pratiques de gestion de ce milieu ne laissent pas celui-ci descendre sous un minimum.

MATÉRIEL ET MÉTHODE

Deux saisons de terrain, 2004 et 2005, ont été consacrées à ce projet de maîtrise. Chacune d'elles correspond à un article distinct, la saison 2004 étant associée à l'article 1 (CHAPITRE 2), la saison 2005 faisant référence à l'article 2 (CHAPITRE 3). Nous présentons successivement les différentes sections pour ces deux saisons.

SAISON 2004

La saison 2004 a été consacrée à la caractérisation de l'habitat, du développement et de la croissance des têtards de grenouille léopard du Nord dans nos deux milieux. À partir du moment où nous avons observé les premières masses d'œufs, soit le 14 mai, jusqu'au début de la métamorphose, le 13 juillet, nous avons visité nos milieux de façon hebdomadaire. À chaque semaine, dans trois stations d'échantillonnage par site, nous mesurions : la profondeur et la température de l'eau, l'azote total (TN) et le phosphore total (TP) (ces données ont été utilisées dans le calcul du rapport azote total : phosphore total (TN :TP)), le coefficient d'extinction de la lumière visible (K_d PAR), la quantité d'algues (Chl *a*) et de matière organique particulaire (POM) disponible dans la colonne d'eau (seston) et sur le substrat végétal (périphyton épiphytique) ainsi que la densité de têtards par mètre cube de colonne d'eau. Ces variables ont été utilisées pour décrire et comparer l'habitat des têtards dans les deux milieux. Nous avons d'abord comparé l'évolution temporelle de ces variables à l'aide de courbes de type *lo(w)ess* (Quinn and Keough 2002), puis réalisé une analyse discriminante pour dégager une fonction qui associerait les stations d'échantillonnage au bon site (naturel ou aménagé), dans une proportion la plus élevée possible.

À chaque semaine, dans les deux milieux et dans les mêmes trois stations d'échantillonnage, des têtards ont été capturés à l'aide de filets troubleaux. Ces têtards ont été ramenés au laboratoire, euthanasiés et conservés dans une solution de formaldéhyde 10% tamponné.

Le stade de développement de chacun des têtards a été déterminé en utilisant la table de Gosner (Gosner 1960). Le poids humide de chacun a également été déterminé. Des courbes de type *lo(w)ess* ont été utilisées pour comparer l'évolution du développement et de la croissance des têtards dans les deux milieux.

Nous avons utilisé un petit bateau gonflable de piscine pour transporter le matériel et les échantillons. Cet outil s'est avéré tellement utile lors de nos deux saisons de terrain que nous avons cru approprié de produire un article pour suggérer à la communauté scientifique de l'utiliser (CHAPITRE 4).

Depuis le début de la métamorphose et ce, durant une semaine, des métamorphes (jeunes grenouilles nouvellement métamorphosées) ont été capturés avec une épuisette. Sur place, ceux-ci étaient mesurés (longueur museau-cloaque : en anglais *snout-vent length* ou SVL) et pesés. Un examen visant à vérifier la présence de malformations et de traces de prédation était aussi réalisé. Lors des travaux de 2004, des données concernant la longueur museau-cloaque (SVL) ont également été prises sur les métamorphes de grenouille léopard provenant de deux autres berges naturelles : Baie-du-Febvre (46°09'N 72°45'O) et Nicolet (46°15'N 72°38'O) et provenant de deux autres aménagements de type bassin permanent : Défense nationale (46°09'N 72°44'O) et Pont Laviolette (46°17'N 72°33') (FIGURE 2.1).

Nous voulions vérifier s'il était possible de généraliser une éventuelle différence de taille à la métamorphose entre les métamorphes provenant des berges naturelles et des aménagements de type bassin permanent. Un test de *t* a été utilisé pour vérifier cette idée.

SAISON 2005

La saison 2005 fut utilisée pour vérifier l'effet de la nature de la ressource alimentaire disponible dans nos deux milieux sur la croissance des têtards. Nous avons donc réalisé une expérience croisée, où des larves de grenouille léopard du Nord provenant de la berge naturelle de Notre-Dame-de-Pierreville ont été placées dans des enclos flottants eux-mêmes placés à la fois dans le site naturel (deux enclos: N3 et N4) et dans le site aménagé (deux enclos: N1 et N2). De la même façon, des larves provenant du site aménagé de la Commune de Baie-du-Febvre ont été placées dans des enclos flottants, à la fois dans le site naturel (deux enclos: A3 et A4) et dans le site aménagé (deux enclos: A1 et A2). Deux masses d'œufs ont été récoltées dans chacun des milieux, naturel et aménagé, puis rapportées au laboratoire où elles ont été élevées dans des bassins individuels. L'eau était oxygénée à l'aide d'une pompe d'aquarium et changée à tous les deux jours. Suite à l'éclosion, deux groupes de 50 larves ont été capturés au hasard dans chacun des bassins, pour un total de huit groupes de 50 larves. Chaque groupe de larves a été placé dans un des enclos flottant le 13 mai 2005. Les enclos avaient été placés dans les milieux le 3 mai 2005, soit dix jours avant que l'on y dépose les larves.

Les enclos flottants étaient essentiellement composés d'une structure de bois rattachée à des flotteurs de styromousse leur permettant de demeurer à la surface. À l'intérieur de la structure de bois était attachée une poche de moustiquaire (maille: 1,25 mm). Les larves ont été placées dans cette poche, profonde de 22 cm. Les têtards ont donc été élevés dans les 22 premiers centimètres, depuis la surface de l'eau. L'ouverture des mailles permettait au seston de circuler librement dans l'enclos. Au fond de la poche, nous avions placé 24 tuiles de grès non-poli qui servaient de substrat pour la croissance du périphyton. Nos enclos fournissaient donc aux têtards, à la fois une source de nourriture à filtrer et une autre source à brouter et correspondait donc davantage à la réalité alimentaire de ceux-ci.

Avant même de pouvoir commencer la caractérisation de l'environnement dans les enclos, les vents violents et les vagues ont détruit les poches de nos enclos dans le milieu naturel (enclos N3, N4, M3 et M4), libérant ainsi les larves. Nous n'avons donc pas pu réaliser l'expérience comme nous le souhaitions. Nous avons tout de même poursuivi nos travaux dans le site aménagé pour obtenir des informations sur le taux de survie, la taille à la métamorphose et l'évolution de la taille des métamorphes durant la période de métamorphose, des têtards provenant des deux origines (enclos N1, N2, M1 et M2).

Une fois par semaine, nous avons donc caractérisé l'environnement des têtards dans chacun des enclos du site aménagé à l'aide des variables suivantes: température de l'eau, pH, oxygène dissous, coefficient d'extinction de la lumière visible, quantités de matière organique en suspension dans la colonne d'eau et sur les tuiles. Les têtards n'ont pas été manipulés durant leur croissance, ainsi on ne peut connaître l'évolution de leur densité au fil du temps.

À partir du moment où un premier métamorphe fut aperçu, la visite des enclos fût quotidienne, jusqu'à la fin de la période de métamorphose. Lorsqu'un métamorphe était aperçu dans un enclos, ce dernier était ramené au laboratoire pour être mesuré (SVL) et pesé. Des métamorphes provenant de chacun des milieux d'origine, naturel et aménagé, ont été capturés, mesurés et pesés pour obtenir des valeurs témoins afin de comparer la taille des métamorphes élevés dans nos enclos.

Une analyse en composantes principales et une analyse discriminante ont été utilisées pour vérifier s'il existait des différences importantes dans l'environnement des têtards dans les quatre enclos au cours de leur développement.

Nous avons vérifié s'il y avait une différence significative dans le poids des métamorphes des différents enclos et des groupes témoins en utilisant un test non-paramétrique de Kruskal-Wallis. Un test post-hoc de Kolmogorov-Smirnov a par la suite été utilisé pour grouper ensemble les métamorphes provenant des enclos et des sites d'origine (groupes témoins), selon leur poids à la métamorphose.

RÉSULTATS ET DISCUSSION

SAISON 2004

L'environnement des têtards durant les premières semaines du suivi était relativemment sembable, soumis à une baisse constante du niveau d'eau. C'est principalement à partir de la sixième semaine que des tendances divergentes se dessinent. Le milieu naturel connaît alors une baisse importante du niveau d'eau, couplée à l'augmentation de la concentration en azote, en phosphore et du coefficient d'extinction de la lumière. Durant les dernières semaines du suivi, les têtards du milieu naturel se retrouvent concentrés dans les dépressions où l'eau est toujours présente. La nourriture est disponible en plus faible quantité et la température de l'eau y est un peu plus chaude. De l'autre côté, le milieu aménagé connaît également une baisse du niveau d'eau mais celui-ci se stabilise durant la deuxième moitié du développement et les têtards ne sont jamais contraints dans des dépressions. On note une augmentation graduelle de la quantité de nourriture disponible, tant sestonique que périphytique. (FIGURE 2.4 à 2.16).

Les variables TN : TP, K_d PAR, Chl *a* sestonique et densité de têtards ont été intégrées dans une fonction discriminante qui associaient les stations d'échantillonnage au bon milieu (naturel ou aménagé) dans une proportion de 75 à 81% (TABLEAU 2.1). En rapportant les différentes stations d'échantillonnage dans une figure présentant les *scores* obtenus par les stations en fonction du temps, on constate que tout au cours du développement, l'environnement des têtards du site aménagé était caractérisé par un ratio TN : TP supérieur, un K_d PAR inférieur, une disponibilité alimentaire sestonique supérieure et une densité de têtards inférieure par rapport à l'environnement des têtards dans le site naturel.

Le développement des têtards, c'est-à-dire la variation du stade Gosner moyen en fonction du temps, s'est déroulé de façon assez similaire tout au cours du suivi.

Cependant, l'écart entre le stade de développement des têtards les moins et les plus avancés dans leur développement, est plus grand dans le milieu naturel que dans le site aménagé, et ce particulièrement lors des dernières semaines du suivi, correspondant au début de la période de métamorphose (FIGURE 2.17).

La croissance des têtards, c'est-à-dire la prise de poids en fonction du temps, fut lente dans les deux milieux durant les trois premières semaines du suivi. Par la suite, les têtards des deux milieux ont amorcé une croissance rapide, les têtards du milieu aménagé présentant toutefois une croissance nettement supérieure à celle des têtards du site naturel qui plafonnent rapidement (FIGURE 2.18). Juste avant le début de la métamorphose, les têtards du site aménagé étaient en fait trois fois plus lourds que ceux du site naturel.

Les métamorphes du site aménagé étaient également plus lourds et plus longs que ceux provenant du site naturel. Un pourcentage plus faible de métamorphes du site aménagé présentait des malformations ou des traces de prédation. Néanmoins, le pourcentage de malformations/traces de prédation dans le milieu naturel était relativement bas, inférieur à 6%. La comparaison de la longueur museau-cloaque (SVL) mesurée chez des métamorphes provenant de d'autres sites naturels et aménagés nous permet d'avancer le fait que la différence de taille observée n'est pas unique à nos deux milieux, mais qu'elle s'observe de façon généralisée sur la rive sud du lac Saint-Pierre.

Notre premier objectif était de vérifier si les milieux humides aménagés de type « bassin permanent » pouvaient permettre aux têtards de grenouille léopard du Nord de compléter leur développement sans anomalies particulières. Les résultats que nous avons obtenus dans un milieu aménagé permanent tendent à confirmer cette hypothèse. Les aménagements permanents, conçus au départ pour permettre à la sauvagine d'élever leur couvée, peuvent donc être considérés comme des milieux « sources » pour la grenouille léopard du Nord, comme l'avaient avancé Schueler et Karstad.

Non seulement les têtards sont en mesure de compléter leur développement dans ces milieux, mais leur croissance y est supérieure. La taille à la métamorphose est un élément clé dans l'histoire de vie des amphibiens (Wilbur et Collins 1973; Smith 1987). Aussi, la taille supérieure des métamorphes des milieux aménagés pourrait leur conférer différents avantages d'ordre écologique et physiologique favorisant leur survie individuelle et leur capacité à coloniser d'autres habitats. Dans l'optique où les métamorphes des sites aménagés n'ont pas moins de chances de survivre que ceux provenant des sites naturels et qu'ils soient ainsi en mesure de coloniser des habitats voisins, la présence des milieux aménagés est d'autant plus intéressante puisqu'elle s'inscrie dans un contexte de métapopulation, vital à la survie des populations d'anoures.

Notre second objectif était de décrire et de comparer l'environnement des têtards dans nos deux milieux humides. La similarité entre nos deux milieux durant les premières semaines du suivi pourrait s'expliquer par la disponibilité d'habitats semblables, soit les marécages arborescent et arbustif. Au fur et mesure que les têtards se développent, ceux du site naturel sont limités au marais, un milieu ouvert, où ils sont concentrés dans un volume réduit. Les têtards du site aménagé demeurent dans le marécage arbustif jusqu'à la métamorphose.

Les variables retenues dans la fonction discriminante, rapport TN : TP, K_d PAR, Chl *a* sestonique et densité de têtards, nous renseignent sur les différences entre nos deux sites. Le rapport TN : TP couplé au coefficient d'extinction de la lumière (K_d PAR), sont responsables en partie de la qualité de la nourriture disponible pour des herbivores comme les têtards puisqu'ils influencent la composition spécifique des communautés d'algues. On sait par exemple qu'un rapport TN : TP inférieur à 29, associé à un coefficient d'extinction de la lumière élevé, favorisera l'émergence de cyanobactéries (Smith 1986; Scheffer *et al.* 1997), lesquelles représentent une nourriture de moins bonne qualité (Brett et Müller-Navarra 1997) comparativement par exemple aux diatomées, considérées comme de la nourriture de bonne qualité (Huggins *et al.* 2004). Suivant ce raisonnement, les conditions environnementales du site naturel devraient avoir favorisé la disponibilité d'une nourriture de moins bonne qualité aux têtards.

On sait que la qualité de la nourriture influence la croissance et la taille à la métamorphose (Kupferberg 1997). La différence dans la qualité de la nourriture disponible dans nos deux milieux pourrait expliquer en partie la différence de taille à la métamorphose observée.

Comme la qualité, la quantité de nourriture disponible pour les têtards influence leur croissance et leur taille à la métamorphose. Les têtards se développant dans les environnements les plus riches se métamorphosant à une taille supérieure (Murray 1990; Johnson 1991). La densité est intimement liée à la quantité de nourriture disponible puisqu'une quantité donnée de nourriture ne représente pas la même chose si quelques ou plusieurs têtards dépendent de cette nourriture. Il est généralement reconnu qu'une densité élevée de têtards dans un milieu conduit à une plus faible croissance et à une plus petite taille à la métamorphose (Gromko *et al.* 1973; John et Fenster 1975; Wilbur 1977; Smith 1983). Nos résultats vont de pair avec ces conclusions puisque les têtards ayant évolué dans l'environnement présentant la plus faible quantité de nourriture est aussi celui qui a produit les métamorphes les plus petits. Les différences dans la quantité de la nourriture disponible ainsi que dans la densité de têtards pourraient expliquer en partie, la différence de taille à la métamorphose.

Les quatre variables retenues dans la fonction discriminante contribuent à notre compréhension de la différence de taille observée chez les têtards et les métamorphes. Nous avançons cependant que la cause proximale de cette différence réside dans le confinement des têtards dans des dépressions durant les dernières semaines du développement. Le niveau d'eau influence la connectivité des surfaces d'habitats : plus haut est le niveau d'eau, plus grande est la connectivité et plus grande est la surface d'habitat disponible pour les organismes strictement aquatiques. Dans un contexte de grande connectivité, les têtards sont en mesure de se disperser et de se nourrir en plus faible densité. Ce scénario prévaut dans nos sites durant les premières phases du développement. C'est d'ailleurs durant cette période que les têtards du site naturel prennent l'essentiel de leur masse puisqu'ils atteignent leur taille de métamorphose à la sixième semaine.

Dans un autre contexte, où la connectivité est faible, la surface disponible est réduite et les têtards sont forcés de s'aggréger. Une variation drastique de la connectivité se produit dans le site naturel entre la cinquième et la sixième semaine. Cette variation se produit uniquement dans le site naturel puisque le site aménagé est indépendant du fleuve Saint-Laurent. La réduction du niveau d'eau a pour conséquence de concentrer les têtards, comme les particules, dans les dépressions. Ces nouvelles conditions environnementales (rapport TN : TP plus bas, K_d PAR élevé, faible quantité de nourriture disponible et densité de têtards élevée), défavorisent la croissance comme nous l'avons discutté précédement. Ces conditions « stressantes » favorisent plutôt le développement des têtards qui cherchent à quitter le milieu aquatique, ce qui a comme conséquence, la production de métamorphes de plus petite taille, tel que suggéré par Wilbur et Collins (1973).

SAISON 2005

La longueur moyenne de la période de développement des têtards, depuis la récolte des masses d'œufs le 5 mai 2005 jusqu'à la fin de la période de métamorphose (période larvaire + métamorphose), a été de 92 jours (n=4; ET : 2,6); alors que la longueur moyenne de la période de métamorphose, c'est-à-dire la période entre l'observation d'un premier métamorphe et du dernier métamorphe, a été de 30 jours (n=4; ET : 3).

Le taux de survie a été très variable d'un enclos à l'autre (TABLEAU 3.1). Les têtards originaires du milieu aménagé ont présenté des écarts importants dans le taux de survie, avec des valeurs de 14 et 66 %. Ceux provenant du site naturel ont présenté des taux similaires et intermédiaires de 36 et 38 %.

Nous ne sommes pas en mesure de connaître l'évolution de la densité de têtards dans chacun des enclos au cours du développement puisque les têtards n'ont pas été manipulés.

Cependant, nous avons déterminé leur densité au début et tout au cours de la période de métamorphose en utilisant les données concernant le retrait quotidien des métamorphes de chacun des enclos. Les résultats démontrent que la densité de têtards dans les différents enclos était inégale (TABLEAU 3.1).

La densité dans les enclos contenant des têtards provenant du milieu naturel était similaire, alors que celle des enclos abritant des têtards du milieu aménagé était opposé (densités faible et élevée) (TABLEAU 3.1).

L'évolution de la densité durant la période de métamorphose (FIGURE 3.3) a présenté un patron similaire dans les enclos abritant des têtards originaires du site naturel : on observe un saut important au début de la métamorphose, alors que 50% des juvéniles émergent dans les trois à six premiers jours, suivi par un plateau durant lequel les individus restants émergent petit à petit. L'enclos M2 (têtards originaires du site aménagé), ayant présenté la densité de têtards la plus faible, ne présente pas de saut particulier mais plutôt une période de métamorphose en plateau, durant laquelle l'émergence de juvéniles se fait tranquillement, de façon espacée dans le temps. Finalement, l'enclos M1 (têtards originaires du site naturel), avec la densité de têtards la plus forte, a présenté un patron de métamorphose caractérisé par deux sauts : un premier durant lequel environ 25 % des têtards survivants ont émergé, suivi par un premier plateau, puis par un second saut durant lequel environ 50% des têtards présents au début de la métamorphose ont émergé. Ce dernier saut a été suivi par un deuxième plateau où les derniers têtards présents se sont métamorphosés petit à petit (FIGURE 3.3).

Nous avons obtenu une différence significative entre le poids des métamorphes provenant des différents enclos, ceux de l'enclos M2 étant significativement (p=0,013) plus lourds que ceux des autres enclos. Nous n'avons cependant pas observé de différence significative concernant le poids à la métamorphose en fonction de l'origine des têtards, soit naturelle ou aménagée (p=0,253).

Le test de Kolmogorov-Smirnov, appliqué aux données de poids des métamorphes provenant des quatre enclos et de ceux capturés dans les milieux d'origine comme témoins, a regroupé ceux-ci en trois groupes. Le premier groupe est composé des plus petits métamorphes des enclos (M1, N1 et N2). Le second groupe est composé des métamorphes de poids intermédiaire de l'enclos M2 et des témoins du site naturel. Le troisième groupe comprend les métamorphes les plus lourds, ceux ayant été capturés dans le site aménagé, mais à l'extérieur des enclos pour servir de témoins (FIGURE 3.3). Finalement, le poids à la métamorphose était négativement associé aux taux de survie des têtards (FIGURE 3.4).

La relation entre le poids à la métamorphose et la durée du développement est positive dans tous nos enclos (FIGURE 3.5). Les individus qui demeurent plus longtemps dans le milieu aquatique croissent davantage et se métamorphosent à une taille supérieure.

Outre la densité de têtards, les variables environnementales n'ont pas montré de différence marquée entre les enclos. Les analyses mutlivariées (en composantes principales et analyse discriminante) utilisées n'ont pas été en mesure de dégager des différences ou des tendances particulières. Aussi, nous considérons que l'environnement dans les différents enclos était semblable tout au long du développement des têtards. Le tableau 3.3 présente les résultats du suivi environnemental dans chacun des enclos.

Les taux de survie observés dans nos enclos (14 à 66%) sont nettement supérieurs à ce que rapporte la littérature en ce qui a trait à la survie en milieu naturel. Le suivi de certaines populations d'anoures nord américaines a en effet montré des taux de survie variant de 1 à 8,5 % (Herreid et Kinney 1966; Calef 1973; Hine *et al.* 1981). Les têtards de nos enclos ont évolué dans un environnement où la pression de prédation devait être plus faible que dans le milieu environnant puisque nous retirions hebdomadairement les prédateurs que nous pouvions repérer. Nos hauts taux de survie ne sont probablement pas étrangers à cette pratique.

C'est probablement la pression de prédation plus élevée dans l'enclos M2 qui y explique le plus faible taux de survie (14%). Nous avons en effet retiré de cet enclos sept larves d'odonates au cours de l'expérimentation, alors que dans les autres nous en avons retiré que une ou deux. De plus, parmi les métamorphes provenant de cet enclos, deux d'entre eux présentaient des traces de prédation : l'amputation d'un ou de deux membres postérieurs. Le très haut taux de survie observé dans l'enclos M1 (66 %) s'explique plus difficilement. Il est possible que la pression de prédation y est été simplement plus faible et qu'ainsi un plus grand nombre d'individus est survécus jusqu'à la métamorphose. Cette hypothèse ne concerne en rien l'origine respective des têtards et fait plutôt référence aux événements qui ont pus influencer le développement des têtards dans un enclos donné. Une autre hypothèse pourrait mettre en relation l'origine des têtards (naturel ou aménagé) et la capacité intrisèque de ceux-ci à exploiter (brouter, filtrer et assimiler) la nourriture disponible. Cette hypothèse suggère donc que les têtards des milieux aménagés exploitent de façon plus efficace les communautés d'algues/protistes des milieux aménagés et que les têtards des milieux naturels exploitent plus efficacement la nourriture disponible dans les sites naturels. Par conséquent, des têtards des milieux naturels déplacés dans un site aménagé, contraint de s'alimenter de la communauté d'algues/protistes propre au site aménagé, auraient moins de chance de survie si leur capacité à exploiter la ressource alimentaire était moindre. Le faible nombre d'enclos utilisé dans cette expérience ne nous permet pas d'avancer une réponse à cette question. La comparaison anatomo-morphologique des têtards pourraient déjà répondre en partie à cette question. Des différences marquées dans l'organisation des pièces buccales (peignes dentaires et papilles) et du tractus digestif (proportion des segments) pourraient signifier une utilisation différente de la ressource alimentaire et par conséquent l'existence de morphotypes intraspécifiques. Cette hypothèse est audacieuse mais il existe de pareils morphotypes chez l'omble de fontaine (*Salvelinus fontinalis*) (Bourke *et al.* 1997), alors pourquoi le phénomène ne pourrait-il pas s'observer chez les têtards d'anoures ?

La relation entre le taux de survie et la taille à la métamorphose est négative. Cette relation suggère un compromis entre la disponibilité de la ressource alimentaire et/ou de l'espace et le nombre et/ou la taille des individus produits par un enclos.

Le taux de survie est en quelque sorte une autre façon d'exprimer la densité dans nos enclos. Une densité élevée est connue chez les anoures pour influencer négativement la croissance en limitant la nourriture disponible par individu (Gromko *et al.* 1973; John and Fenster 1975; Wilbur 1977; Smith 1983). En considérant qu'il n'y avait pas de différence marquée dans la disponibilité de la nourriture des différents enclos, on peut penser qu'effectivement, les têtards devaient compétitionner pour une quantité équivalente de nourriture, tout en étant plus ou moins nombreux. La compétition à l'intérieur des enclos et donc la densité, pourrait expliquer les différences morphologiques observées.

La métamorphose est régulièrement présentée comme une brève période durant laquelle tous les têtards émergent. Nos observations montrent plutôt que dans certaines situations, la métamorphose est un processus relativement long (30 jours dans notre cas), durant lequel les conditions environnementales évoluent et influencent les têtards toujours présents dans le milieu aquatique. Selon Wilbur et Collins (1973), la diminution des stress environnementaux au cours de la métamorphose permettrait aux têtards toujours présents de croître plus rapidement et de se métamorphoser à une taille supérieure. Dans notre suivi, il semble que la réduction graduelle de la densité de têtards dans les enclos, au fur et à mesure que des individus quittaient, ait favorisé la croissance des autres puisque ceux-ci se sont métamorphosés à une taille supérieure.

CONCLUSION

Nos travaux sur le développement et la croissance des têtards de grenouille léopard du Nord dans la plaine d'inondation du lac Saint-Pierre nous amènent à différentes conclusions. D'abord, l'aménagement de grands marais artificiels ayant pour principal objectif de fournir aux canards un habitat de choix pour l'élevage des couvées semble profitable à la grenouille léopard du Nord. Les adultes retrouvent des sites pour pondre tandis que les têtards trouvent des microhabitats propices au développement, ces derniers y connaissant une croissance importante. Le suivi d'une cohorte au fil de son développement a démontré que celle-ci pouvait produire un recrutement de qualité, vraisemblablement en mesure de contribuer au maintien local de l'espèce. L'hypothèse avancée par Schueler et Karstad est vraisemblablement vérifiée à l'échelle du site aménagé. En contrepartie, le rôle des sites aménagés dans l'exportation de juvéniles vers les milieux humides avoisinants demeure inconnu. Il s'agirait là, selon nous, d'une prochaine étape prioritaire dans le contexte de l'étude du rôle écologique des sites aménagés en relation avec les communautés d'anoures.

Le modèle de conception et de gestion de ces aménagements devrait être plus amplement analysé et décrit de manière à dégager les particularités qui favorisent d'abord la grenouille léopard, travail que nous avons amorcé et qui attend d'être complété, mais aussi toute la communauté d'anoures qui gravitent autour et utilisent les sites aménagés. La localisation et la description macroscopique des méso-habitats devraient être réalisées de manière à construire des IQH propres à chaque espèce d'anoures utilisant les sites artificiels du PNAGS. Ces IQH intègreraient non seulement le concept d'habitat mais également celui des pratiques de gestion de ces marais. En effet, les diverses pratiques associées aux sites aménagés, que se soit la vidange de haltes migratoires, tout comme le maintien artificiel du niveau de l'eau, influencent directement le développement des têtards mais probablement aussi la composition spécifique des communautés d'anoures.

Certains ajustements simples de synchronisation de ces pratiques pourraient permettre par exemple, la nidification de la sauvagine mais aussi la reproduction de la grenouille léopard du Nord au printemps, puis de la grenouille verte (*Rana clamitans*) et du ouaouaron (*Rana catesbeiana*) au début de l'été. Ces deux dernières espèces sont présentes dans la plaine inondable du lac Saint-Pierre mais étaient absentes des sites aménagés en 2004. Les besoins de chacune des espèces en termes d'habitats des têtards demeurent à peu près inconnus, alors qu'il s'agit de stades vitaux fortement influencés par les conditions environnementales.

Le suivi d'une cohorte en milieu naturel nous a permis de mieux comprendre l'effet de ces conditions environnementales sur le développement et la croissance des têtards. On a vu par exemple qu'un site, avec des conditions qui défavorisent la qualité et la quantité de la nourriture disponible pour les têtards, semble influencer négativement la croissance individuelle de ceux-ci. Le bas niveau d'eau du Saint-Laurent pourrait être responsable de l'apparition de conditions plus difficiles pour les têtards du site naturel, alors que ceux-ci sont contraints dans des dépressions où se dégrade l'environnement. Nous faisons le parallèle entre cette concentration d'individus en milieu naturel et nos observations provenant de notre étude en enclos. Dans les deux cas, la densité a joué un rôle sur le développement et la croissance des têtards et la métamorphose a perduré. Ces éléments de connaissance contribuent à notre compréhension de l'écologie du développement chez la grenouille léopard du Nord. En relation avec les changements annuels et stochastiques du niveau d'eau du fleuve Saint-Laurent, la compréhension de l'écologie des têtards de cette espèce pourrait être un élément clé lors d'éventuels efforts pour sauvegarder régionalement l'espèce.

BIBLIOGRAPHIE

1. Andrén, C., L. Hendrickson, M. Olsson, and G. Nilson. (1988) Effects of pH and aluminum on embryonic and early larval stages of Swedish brown frogs, *Rana arvalis, R. temporaria and R. dalmatina*. Holarctic Ecology. 11 : 127-135.

2. Benoît, J., R. Bergeron, J.-C. Bourgeois, S. Desjardins, et J. Picard. (1987) Les habitats et la faune de la région du lac Saint-Pierre : synthèse des connaissances. Ministère du Loisir, de la Chasse et de la Pêche du Québec. Directions régionales de Montréal et de Trois-Rivières. Ix + 123 pages.

3. Berven, K. A., and B. G. Chadra. (1988) The relationship among egg size, density and food level on larval development in the wood frog (*Rana sylvatica*). Oecologia. 75 : 67-72.

4. Bourke, P., P. Magnan, and M. Rodriguez. (1997) Individual variations in habitat use and morphology in brook charr. Journal of fish biology. 51: 783-794.

5. Bradford, D. F., and R. S. Seymour. (1988) Influence of environmental PO2 on embryonic oxygen consumption, rate of development, and hatching in the frog, *Pseudophryne bibroni*. Physiological zoology. 61 : 470-474.

6. Calef, G.W. (1973) Natural mortality of tadpoles in a population of *Rana aurora*. Ecology 54 : 741-758

7. Crowder, W. C., M. Nie, G. R. Ultsch. (1998) Oxygen uptake in bullfrog tadpoles (*Rana catesbeiana*). Journal of experimental zoology. 280 : 121-134.

8. Cummins, C. P. (1989) Interaction between the effects of pH and density on growth and development in *Rana temporaria* tadpoles. Functional ecology. 3 : 45-52.

9. Daigle, C., et J. Jutras. (2001) Récolte d'anoures au Québec en 1998. Société de la faune et des parcs du Québec. 14 pages.

10. Denver, R. J., N. Mirhadi, and M. Phillips. (1988) Adaptative plasticity in amphibian metamorphosis : response of *Scaphiopus hammondii* tadpoles to habitat dessication. Ecology. 79 : 1859-1872.

11. Desroches, J.-F., et D. Rodrigue. (2004) Amphibiens et reptiles du Québec et des Maritimes. Éditions Michel Quintin. Waterloo, Canada. 288 p.

12. Desroches, J.-F., F. W. Schueler, I. Picard, and L.-P. Gagnon. (in press) A herpetological survey of the James Bay area of Québec and Ontario. Submitted to Canadian Field-Naturalist.

13. D.U.C. (Ducks Unlimited Canada), et N.O.V.E. environnement inc. (1990) Projet d'aménagement de la Commune de la Baie-du-Febvre. Étude d'impact sur l'environnement déposée au près du Ministère de l'Environnement du Québec. 134 pages.

14. FAPAQ (Société de la Faune et des Parcs du Québec). (2003) Le lac Saint-Pierre : Portrait des aménagements et autres sites d'intérêt faunique. Direction de l'aménagement de la faune Mauricie – Centre-du-Québec, Trois-Rivières, Canada. 176 pages.

15. Gilbert, M., R. Leclair Jr., and R. Fortin. (1994) Reproduction of the northern leopard frog (*Rana pipiens*) in floodplain habitat in the Richelieu river, P. Quebec, Canada. Journal of Herpetology 28 :465-470.

16. Gosner, K. L. (1960) A simplified table for staging anuran embryos and larvae with notes on identification. Herpetologica 16 :183-190.

17. Gromko, M. H., F. S. Mason, and S. J. Smith-Gill. (1973) Analysis of the crowding effect in *Rana pipiens* tadpoles. Journal of experimental zoology. 186 : 63-72.

18. Harkey, G. A., and R. D. Semlitsch. (1988) Effects of temperature on growth, development and color polymorphism in the ornate chorus frog *Pseudacris ornata*. Copeia 4 : 1001-1007.

19. Herreid II, C. F., and S. Kinney. (1967) Temperature and development of the wood frog *Rana sylvatica*, in Alaska. Ecology. 48 : 579-590.

20. Hine, R.L., B.L. Les and, B.F. Hellmich. (1981) Leopard frog populations and mortality in Wisconsin, 1974-76. Technical Bulletin No. 122. Department of natural resources. Madison, Wisconsin. 38 pages.

21. Huggins, K., J.-J. Frenette, and M. T. Arts. (2004) Nutritional quality of biofilms with respect to light regime in Lake Saint-Pierre (Québec, Canada). Freshwater Biology 49: 945–959.

22. John, K. R., and D. Fenster. (1975) The effects of partitions on the growth rates of crowded *Rana pipiens* tadpoles. American Naturalist. 93:123-130.

23. Johnson, L. M. (1991) Growth and development of larval northern cricket frog (*Acris crepitans*) in relation to phytoplankton abundance. Freshwater biology. 25 :51-59.

24. Kupferberg, S. (1997) The role of larval diet in anuran metamorphosis. American zoologist. 37 : 146-158.

25. Langlois, C., L. Lapierre, M. Léveillé, P. Turgeon, et C. Ménard. (1992) Synthèse des connaissances sur les communautés biologiques du lac Saint-Pierre. Rapport technique, Zone d'intérêt prioritaire no 11. Centre Saint-Laurent, Environnement Canada. 236 pages.

26. Murray, D. L. (1990) The effects of food and density on growth and metamorphosis in larval wood frogs (*Rana sylvatica*) from central Labrador. Canadian journal of zoology. 68 : 1221-1228.

27. (NAWMP) North American Waterfowl Management Plan, Plan Committee. (2004) North American Waterfowl Management Plan 2004. Strategic guidance: strengthening the biological foundation. Canadian Wildlife Service, U.S. Fish and Wildlife Service, Secretaria de Medio Ambiente y Recursos Naturales. 22 p.

28. Quinn, G. P., and M. J. Keough. (2002) Experimental Design and Data Analysis for Biologists. Cambridge University Press. Cambridge, UK. 537 pages.

29. Scheffer, M., S. Rinaldi, A. Gragnani, L. R. Mur, and E. H. Nes. (1997) On the dominance of filamentous cyanobacteria in shallow, turbid lakes. Ecology. 78: 272-282.

30. Schueler, F. W. and A. Karstad. (2000) Northern Ontario Ducks Unlimited marshes: refugia for leopard frogs? Proceeding of the 5th Canadian amphibian and reptile conservation network annual meeting.

31. Smith, D. C. (1983) Factor controlling tadpole populations of the chorus frog (*Pseudacris triseriata*) on Isle Royale Michigan. Ecology. 64: 501-510.

32. Smith, D. C. (1987) Adult recruitment in chorus frogs: effects of size and date at metamorphosis. Ecology 68:344-350.

33. Smith, V. H. (1986) Light and nutrient effects on the relative biomass of blue-green algae in lake phytoplankton. Canadian Journal of Fisheries and Aquatic Sciences. 43:148-153.

34. Steinwascher, K. (1978) The effect of coprophagy on the growth of *Rana catesbeiana* tadpoles. Copeia. 1 : 130-134.

35. Wilbur, H. M. (1977) Density-dependent aspects of growth and metamorphosis in *Bufo americanus*. Ecology. 58 : 196-200.

36. Wilbur, H. M., and J.P. Collins. (1973) Ecological aspects of amphibian metamorphosis. Science. 182: 1305-1314.

CHAPITRE 2

ARTICLE 1

North American Waterfowl Management Plan Permanent Basin as an Effective Tool for Northern Leopard Frog (*Rana pipiens*) Conservation

Submitted to Conservation Biology

North American Waterfowl Management Plan Permanent Basin as an Effective Tool for Northern Leopard Frog (*Rana pipiens*) Conservation

DANIEL POULIOT, and JEAN-JACQUES FRENETTE
Département de chimie-biologie, Université du Québec à Trois-Rivières, 3351 boulevard des Forges, C.P. 500, Trois-Rivières, Québec, Canada G9A 5H7

Abstract: We characterized the environment, development and growth of the northern leopard frog's (*Rana pipiens*) tadpoles in two sites: a natural bay of the St. Lawrence River and a permanent basin managed for waterfowl in the context of the North American Waterfowl Management Plan (NAWMP). We were interested in investigating the hypothesis that permanent managed wetland could be considered as "source habitat" for this species. Environmental conditions presented similarities but it seems that tadpoles from the permanent basin developed in more productive environment and in lower density. Patterns of water level fluctuations seem to be the chief cause of those environmental differences. Tadpoles from the permanent basin grew more and metamorphs were about two times heavier than those from the natural bay. Size differences were still significant when comparing metamorphs from other similar sites. NAWMP permanent basin can be considered as a "source habitat" for the northern leopard frog and appears to be a useful tool for the conservation of this declining species.

Keywords: North American Waterfowl Management Plan, Permanent basin, Northern leopard frog, *Rana pipiens*, tadpoles, food quality, food quantity, density, hydrology.

Introduction

The North American Waterfowl Management Plan (NAWMP) was signed by Canada and USA in 1986, in response to major waterfowl decline observed throughout North America; Mexico joined the continental plan in 1994 (NAWMP Plan Committee 1998). The main goal of this plan is to return duck and goose populations to their 1970's level (NAWMP Plan Committee 2004). Habitat losses and wetland degradation over the last decades have greatly contributed to the waterfowl decline (Batt *et al.* 1989). Amongst the solutions used by the NAWMP partners to achieve their goal was the establishment of a network of managed and/or protected wetlands across Canada, USA and Mexico. NAWMP aimed to diversify the type of wetlands in response to the specific needs of waterfowl (D.U.C. and NOVE 1990). Some of the managed wetlands are temporary i.e. land is flooded in early spring and acts as a migratory stop; the land is then drained at the beginning of the summer to allow for agriculture (FAPAQ 2003). Other managed wetlands are permanent and designed mainly for the nesting and the raising of juveniles by waterfowl (D.U.C and NOVE 1990).

The NAWMP wetlands have initially been developed for waterfowl, however, managers soon realised the great potential of the managed areas for biological conservation. Some "managed wetland" success stories have been recently published and emphasized the quality of those wetlands for a great diversity of birds (Locky *et al.* 2005; VanRees-Siewert and Dinsmore 1996) and for the growth of fish (e.g. yellow-perch (*Perca flavescens*)) (Tardif *et al.* 2005). Moreover, Schueler and Karstad (2000) mentioned the high abundance of the northern leopard frog (*Rana pipiens*) in managed wetlands of northern Ottawa River drainage in Ontario, Canada. They proposed that those wetlands may act as refuges or "source habitats" for that species and could contribute to the species survival at the local scale. In this case, a "source habitat" refers to a habitat which allows the tadpoles to achieve their development without major abnormalities.

Although the northern leopard frog (*Rana pipiens*) is not considered "at risk" at the continental scale, a significant decline has been observed over most of its range during the last 30 years (Seburn and Seburn 1998; Gilbert *et al.* 1994; Wassersug 1976; Gibbs 1971). Populations from western Canada are now considered "endangered" (Southern mountain populations) and of "special concern" (Western boreal/Prairie populations) (COSEPAC 2002). Canadian eastern populations are still abundant (COSEPAC 2002) but Desroches *et al.* (in press.) documented a serious decline in the James Bay region of north eastern Ontario since 1971. Moreover, they found the species in very few localities during a recent (2002) survey in the Quebec portion of the James Bay.

Many causes can explain the decline of the northern leopard frog (COSEPAC 2002) but the lost or deterioration of habitats, especially the modification of the breeding habitat hydrology, is considered a major cause of decline for that species (Seburn and Seburn 1998; Gilbert *et al.* 1994).

Considering that the NAWMP permanent wetlands have been developed to meet the waterfowl needs and management policies for ducks/geese, the objectives of that study were: a) to consider the possibility for these habitats to act as "source habitats" for northern leopard frogs and b) to compare the tadpoles' habitat in managed and natural wetlands. Such knowledge will contribute to explain the large occurrence of the northern leopard frog in the NAWMP permanent wetlands.

Methods

Study sites

We conducted our research in Lac Saint-Pierre (46°12'N; 72°50'W), the largest fluvial lake (480 km^2) along the St. Lawrence River, Canada. (Fig. 1). Lac Saint-Pierre is characterized by its large floodplain (180 km^2) which includes many natural and NAWMP managed wetlands (FAPAQ 2003). The fluvial lake is shallow (mean depth of 3.17 m during the period of mean discharge) and covered with extensive macrophyte beds during most of the ice-free period.

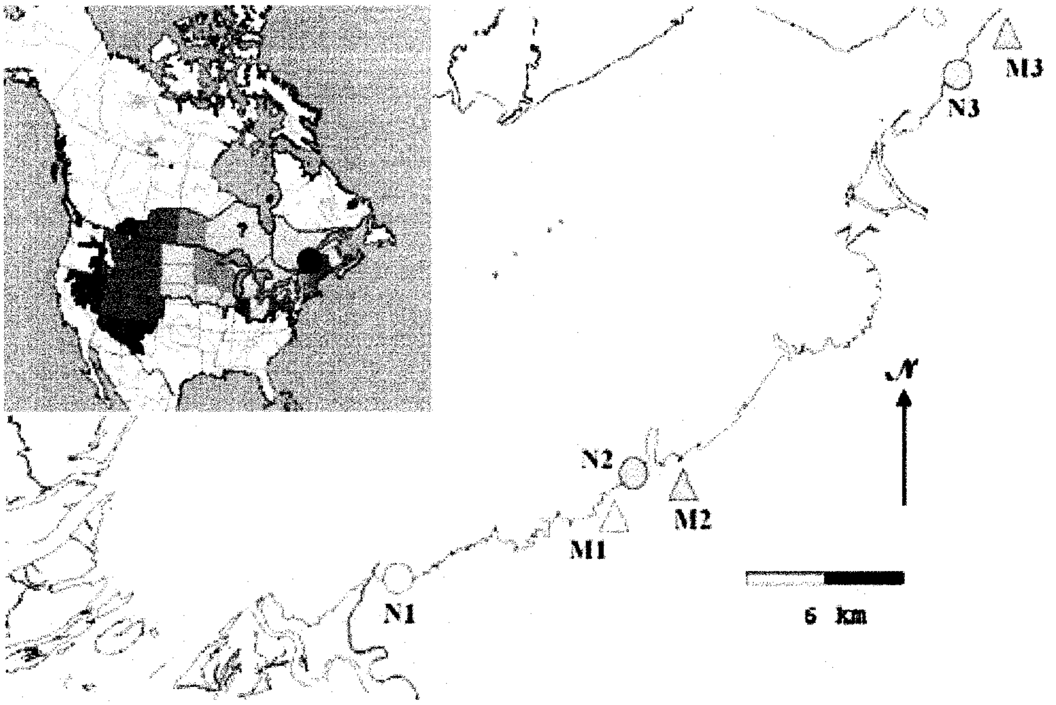


Figure 1. Top-left corner: the circle localized the study area. The northern leopard frog distribution is show in tones of grey following the subnational "S" rank of NatureServe Conservation Status; darker the tone more endangered the northern leopard frog is (S5 to S1). The "?" suggest that status in northern Ontario can be more critical than taught, as suggest by Desroches et al. (in press). Main image: localisation of the study sites in Lac Saint-Pierre, St. Lawrence River, Canada. N1 and M1 represent locations of natural (N) and managed (M) wetlands where tadpoles' environment, development and growth have been characterized. N2, N3, M2 and M3 refer to stations where only snout-vent length of metamorphs was measured.

The natural wetlands of Lac Saint-Pierre are colonized by plant communities which are sequentially unflooded from spring to summer. Water gradually uncovers the silver maple (*Acer saccharinum*) forest characterized by reed canarygrass (*Phalaris arundinacea*) in open areas, and during the later stage, the low depth marsh with dominant common spikerush (*Eleocharis smallii*) and various bulrush species (*Scirpus fluviatilis, S. americanus and S. acutus*). Frog's eggs are laid in the flooded forest but tadpoles have to follow the water mass and their environmental conditions are dictated by the Saint-Lawrence River's water level fluctuations.

Four dikes give the rectangular shape which delimits typical NAWMP permanent managed wetlands (Fig. 3). Water accumulates in these wetlands following snow melting and spring rains.

Water level decreases gradually over the summer by evaporation and transpiration but a pumping station can be activated to add water from the Saint-Lawrence River if it goes too low. Drainage channels are present but the wetland is not usually flushed. They exhibit a depth gradient that allows for the establishment of three aquatic plant communities: (1) the shallow water is dominated by an arborescent swamp of willows (*Salix sp.*) and/or ashes (*Fraxinus sp.*), with dense aggregations of cattail (*Typha angustifolia*); (2) the open marsh is densely colonised by common spikerush and invasive flowering-rush (*Butomus umbellatus*) at mid depth; and (3) open water is covered with submerged vegetation in the deepest areas. Northern leopard frog's eggs are laid both in the arborescent swamp and in the open marsh.

We characterized the tadpole's environment, development and growth weekly, in one natural site (N1) and one permanent managed wetland (M1) during the 2004 season (Fig. 1). The monitoring began with the first observation of egg mass (May 14) and ended with the beginning of the metamorphosis (July 13). In the "Results" section, each week was given a number in order to simplify the figures: Week 1: May 14, Week 2: May 18, Week 3: May 25, Week 4: June 02, Week 5: June 08, Week 6: June 15, Week 7: June 22, Week 8: June 29, Week 9: July 08, Week 10: July 13.

Figure 2. Natural wetland in Lac Saint-Pierre. At mid-summer, the water level decreases in the St. Lawrence River and tadpoles are constrained to small pools like the one shown here. Photo by Daniel Pouliot.

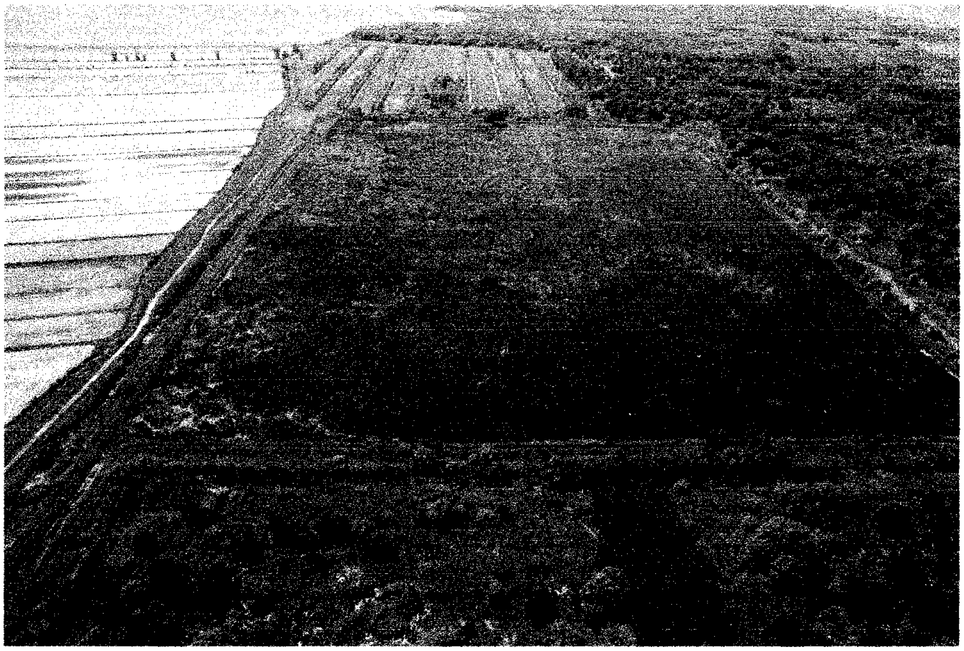


Figure 3. The NAWMP permanent managed wetland where tadpoles' environment, development and growth have been characterized. It exhibits the typical rectangular shape of a permanent NAWMP basin. Photo by André Michaud (Ducks Unlimited Canada). Used with permission.

Localisation of tadpoles' habitat

Studied wetlands are highly heterogeneous ecosystems, as previously described, and tadpoles don't necessarily use all available areas. To ensure that we characterize environmental conditions of areas really occupied by tadpoles, we first localized tadpoles' habitats. Tadpole density (tadpoles/m^3) was measured with a dipnet passed two time in the water column (we followed the bottom, horizontally, for about meter). Filtered water varied from 0.13m^3 to 0.08 m^3 as a function of water depth. Tadpole density was expressed by unit of volume (tadpoles/m^3). Tadpole density was measured at 30 locations randomly selected and minimally separated by 20 meters. The 3 locations with the highest northern leopard frog tadpole density were considered representative of the space occupied by the tadpoles and were designated as "tadpole habitat". We characterized the environment and captured tadpoles in those 3 tadpole habitats to evaluate their development stage and weight. The same sampling strategy was repeated weekly, in both sites.

Environmental conditions

In each of the 3 tadpoles' habitats at both sites, we measured the water depth using a meter stick graduated in centimeter (0.5 cm) and the water temperature with a thermometer (0.5 °C). Photosynthetically active radiation (PAR) was measured in situ with a light meter (Li-COR µmoles/m^2/sec) and extinction coefficient (K_d PAR) calculated following Wetzel and Likens (1991). Four litres of water were collected in 2 L polypropylene containers and prefiltered on 63 µm Nitex (Filmar inc.) to eliminate large zooplankton and minimize grazing on the algae/protista community during the shipment of the water to the lab. Prefiltered water samples were kept in a cooler with ice packs until their arrival at the lab where they were rapidly transferred in the dark at 4°C. A 200 ml subsample was used for total nitrogen and total phosphorus analyses according to standard methods in use at Yves Prairies laboratory (GRIL). For sestonic chlorophyll (Chl *a*) determination, water samples were filtered the same day on duplicate GF/F filter (0.7 µm); extraction was by hot ethanol following Nusch (1980). Measurement of sestonic organic matter dry weight was made according to Wetzel and Likens (1991). These variables were used to characterize the food availability in the water column.

Chl *a* represents a measure of phytoplankton biomass. The sestonic organic matter dry weight includes living organisms and decaying matter as faeces, which are known to be beneficial for tadpoles' development and growth (Steinwascher 1978).

Epiphytic periphyton attached to dominant macrophyte species was used as another index of food availability for tadpoles. Ten grams of submerged parts from the most representative macrophytes were collected in each station in a plastic bag containing 100 ml of distilled water. Bags were agitated twice a minute to thoroughly separate epiphytic periphyton from macrophyte substrate. The result of the washing-shaking process was a volume-known homogenate of distilled water and epiphytic algae/protista. This homogenate was filtered the same day on GF/F filter (0.7 µm) to evaluate the epiphytic Chl *a* biomass and epiphytic organic matter dry weight. Epiphyte Chl *a* and organic matter availability were expressed per unit of plant area, which was measured using an area-meter (AMC100 ADC Bioscientific LTD). Finally, to obtain a more realistic idea of the amount of available epiphytic food resource (Chl *a* and organic matter), we calculated and compared the following epiphytic food availability index (EFA index) for each station:

$$\begin{gathered}\textbf{EFA index} = \underline{\boldsymbol{\mu}\textbf{g Chl}\boldsymbol{a}\textbf{/cm}^{2}} \textbf{ or } \underline{\textbf{mg Organic matter /cm}^{2}} \; * \\ \textbf{Macrophyte percent cover (0-100)} * \textbf{Water depth}\end{gathered}$$

Tadpole density data were obtained from the localisation of tadpoles' habitat as previously described in the subsection: Localisation of tadpole habitats.

Tadpoles' development and growth

Tadpole's development and growth were characterized weekly from tadpoles captured in the 3 tadpoles' habitat of both sites; tadpoles were collected and brought to the lab in a cooler with ice packs. Tadpoles were euthanized using MS-222 and placed in formaldehyde preservative solution (10 % neutral buffered) according to McDiarmid and Alford (1999). For all preserved tadpoles, the development stage was determined using the Gosner table (Gosner 1960).

Tadpoles were individually placed on a dry towel for a few seconds to absorb excessive formaldehyde solution (tadpoles were never dried) and weighed to the nearest 0.05 mg using a Sartorius balance (model CP225D). Shrinking curves such as those proposed by Fisher *et al.* (1998) were not used since we found no particular difference between preserved and fresh samples: tadpoles of all stages were normally turgescent and did not present any major skin folds.

Metamorphs

The week following the beginning of the metamorphosis, we evaluated the length of the metamorphs (Gosner stages 46) in our two initial sites (N1 and M1) and in four others sites (N2, N3 and M2, M3), for a total of six different sites (three natural sites and three permanent managed sites) (Fig. 1). These additional sites were sampled to take into account the effect of environmental differences on the size of the metamorphs produced by different natural and permanent managed wetlands of the Lac Saint-Pierre south shore. Specimens were collected with a dipnet at the water-land interface. Metamorphs were measured for snout-vent length (SVL) with a caliper (Mitutoyo), and examined for malformations or predation signs. Available records from previous years were included in our data set to compare the size at metamorphosis of young frog from natural and permanent managed wetlands.

Statistical Analysis

A graphic analysis of the environment evolution, using *lo(w)ess* curves was firstly done to look for general tendencies, similarities and dissimilarities between the evolution of tadpoles' environment in the two sites (N1 and M1). A discriminant function was obtained following a stepwise DFA (F to enter or reject 0.15) on the environmental variables. This analysis was used to identify variables or groups of variables than can best differentiate the characterized wetlands. A two-tailed t test was used to determine whether there is a significant difference ($\alpha = 0.05$) between the mean length of metamorphs from the natural and managed sites (N1, N2, N3 vs. M1, M2, M3).

Results

Environmental conditions

Mean depth remained consistently high at both sites during the first part of development, from week 1 to 4 (May 14 to June 2). After the fourth week, depth was generally lower in the natural than in the managed wetland (Fig. 4).

Available area for tadpoles to disperse is not expressed by the water depth variable. In some situations, water depth can be equal or similar in two sites but the available area is clearly different (Fig.5). During the first half of the tadpoles' development (week 1 to 5; May 14 to June 8), major hydrological connectivity between the different plant communities let the tadpoles disperse in both sites. But during the second half (from week 6 to 10; June 15 to July 13), dispersion is possible only to tadpoles in the managed wetland as those in the natural wetland were gradually confined to pools formed as the water retreated (Fig. 5).

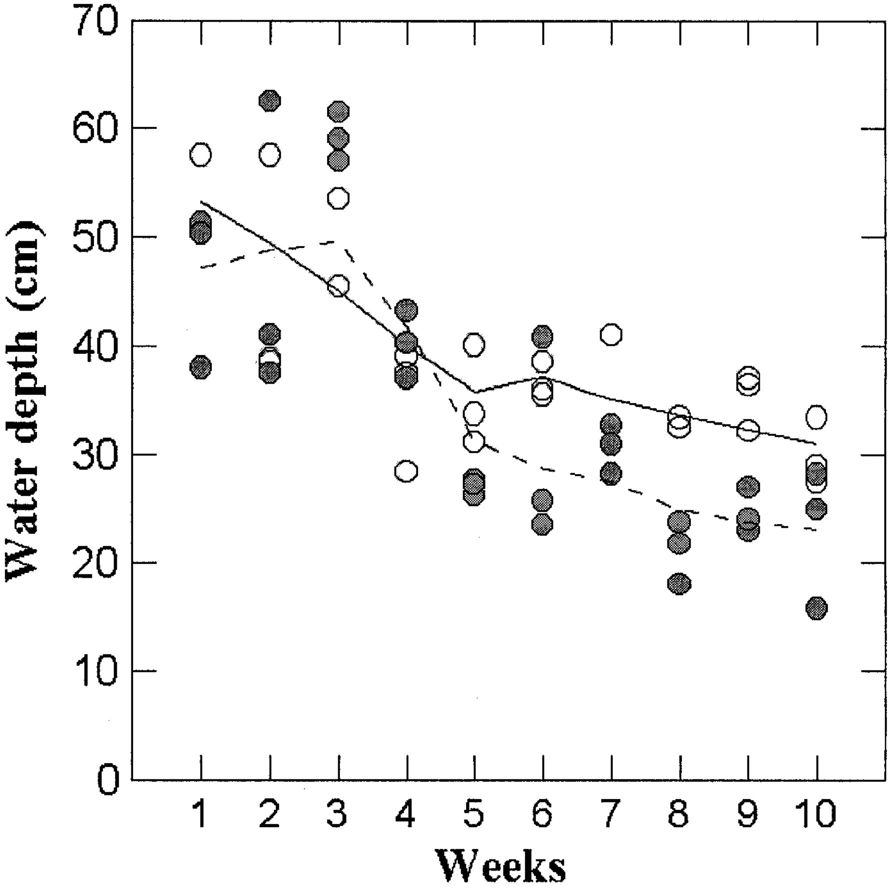


Figure 4. Weekly water depth, from May 14 to July 13, in the tadpoles' habitat of the natural wetland (● and break lo(w)ess) and of the permanent managed wetland (○ and plain lo(w)ess). Smoothing parameter: 0.5.

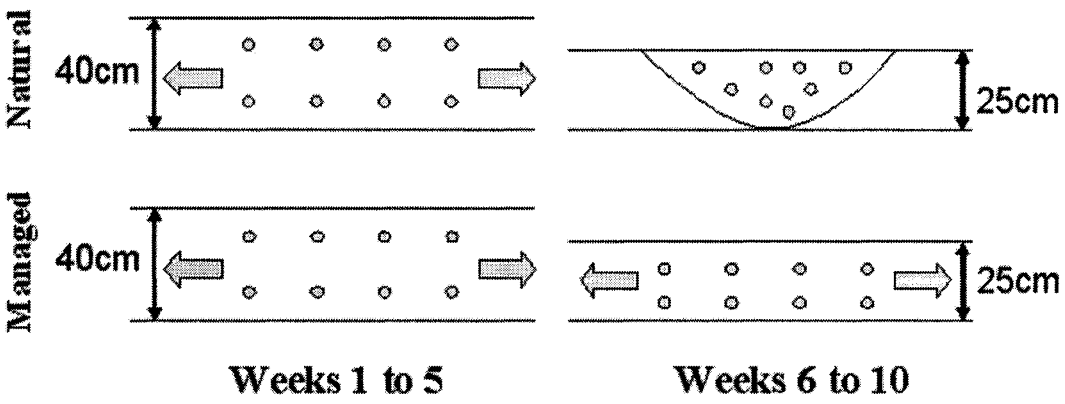


Figure 5. Schematic of spatial habitat availability to tadpoles during the first (weeks 1 to 5: May 14 to June 8) and the second (weeks 6 to 10: June 15 to July 13) half of the development in the natural and managed wetlands. Dots represent tadpoles and lateral arrows, the possibility of dispersal to connecting habitat (their absence indicates that tadpoles are confined to the given area) and vertical arrows, the measured water depth.

Water temperature was variable over time, with warmer conditions in both sites at mid-development (Week 6: June 15). However, water temperature in the natural wetland remains higher until metamorphosis (Fig. 6). Tadpoles from the natural wetland were thus encountered in shallower and warmer locations than those from the permanent managed wetland, particularly during the last three weeks of development (June 29 to July 13).

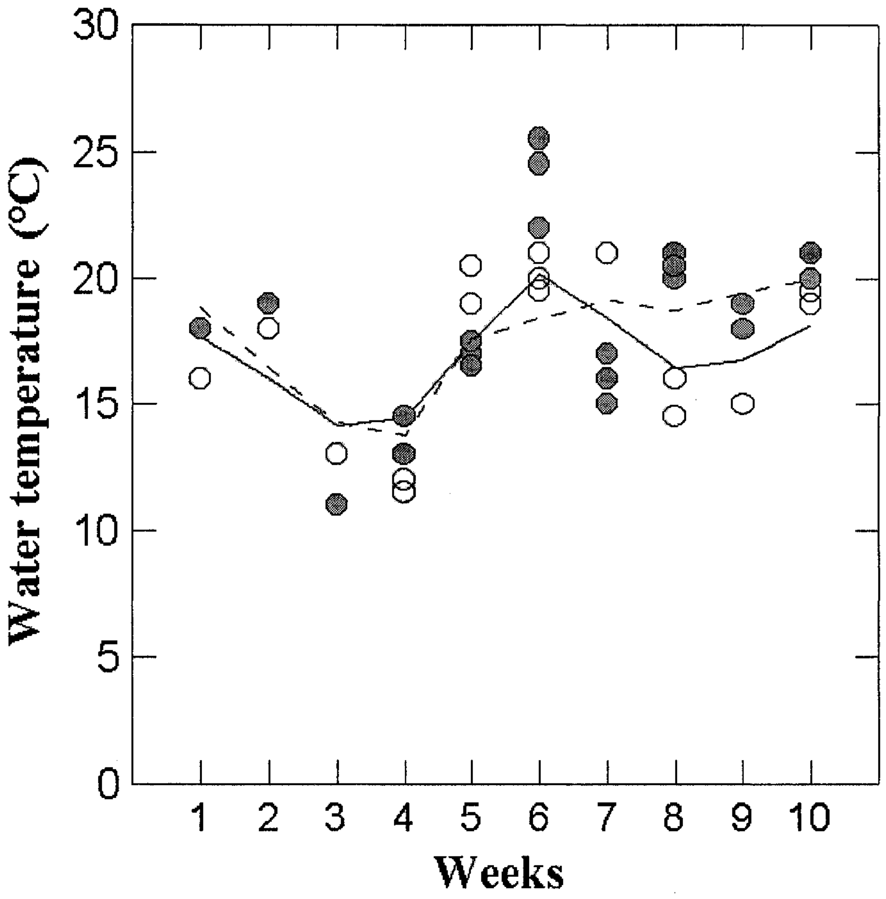


Figure 6. Weekly water temperature, from May 14 to July 13, in the tadpoles' habitat of the natural wetland (● and break lo(w)ess) and of the permanent managed wetland (○ and plain lo(w)ess). Smoothing parameter: 0.5.

Light coefficient extinction (K_d PAR) values were generally similar in both sites at the beginning and at the end of tadpoles' development (Fig. 7). However, K_d PAR values increased by a factor of 2 to 4 in the natural site during mid-development between weeks 5 and 7 (June 8 to 22). During this particular period, mean extinction coefficient was 10.4 (SD: 7.5) in the natural site and 4.5 (SD: 2.0) in the managed wetland.

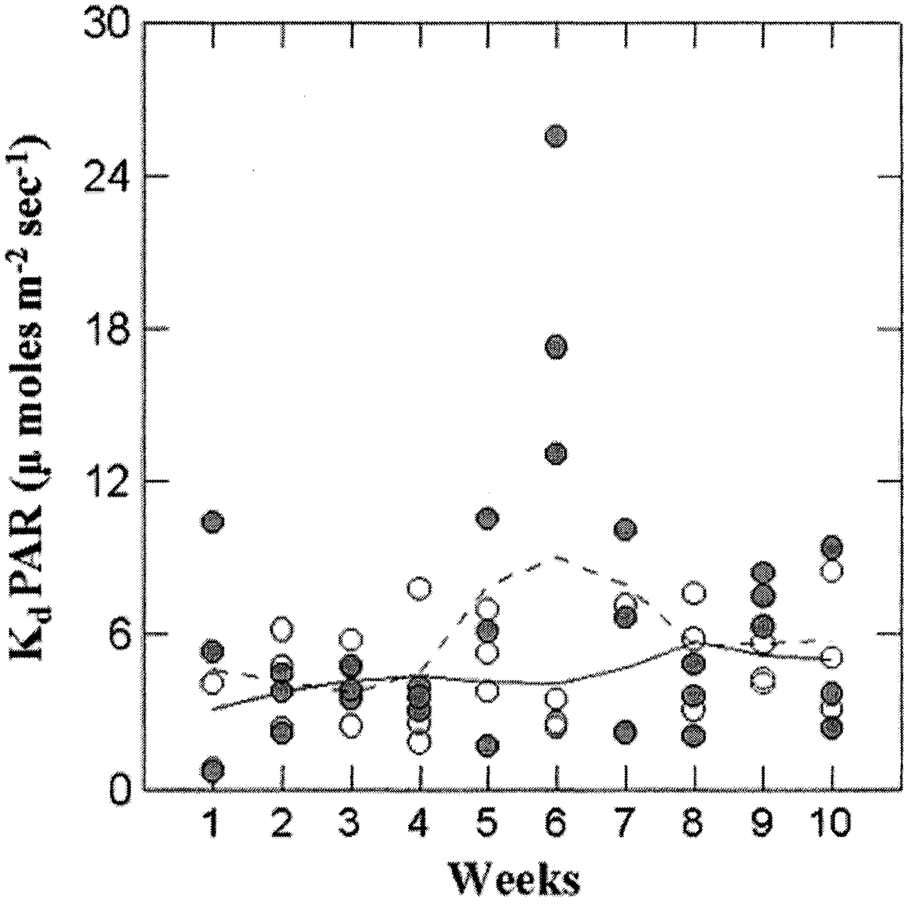


Figure 7. Weekly photosynthetically active radiation extinction coefficient (K_d PAR), from May 14 to July 13, in the tadpoles' habitat of the natural wetland (● and break lo(w)ess) and of the permanent managed wetland (○ and plain lo(w)ess). Smoothing parameter: 0.5.

Total nitrogen concentrations (Fig. 8) were generally higher in the managed wetland than in the natural site during most of the study period. However, as in the case of light extinction coefficient, the natural site has shown outlier points for total nitrogen during mid-development, specifically at week 6 (June 15). An opposite pattern appeared for phosphorus where the natural site was globally richer in this element (Fig. 9).

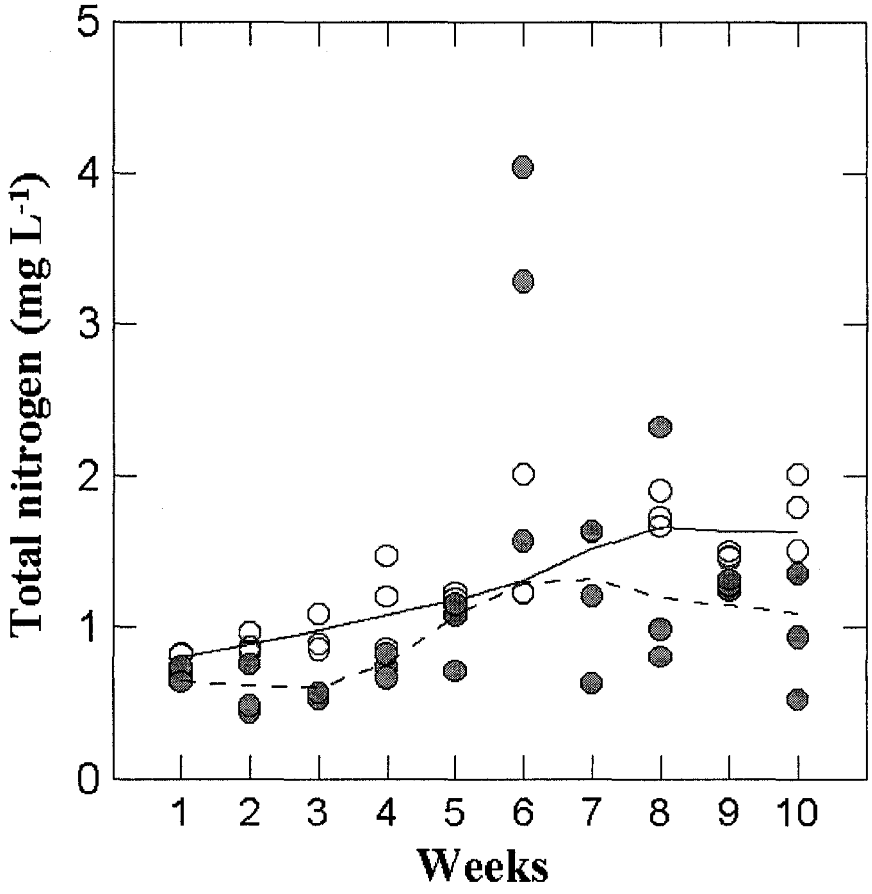


Figure 8. Weekly total nitrogen, from May 14 to July 13, in the tadpoles' habitat of a natural wetland (● and break lo(w)ess) and a permanent managed wetland (○ and plain lo(w)ess). Smoothing parameter: 0.5.

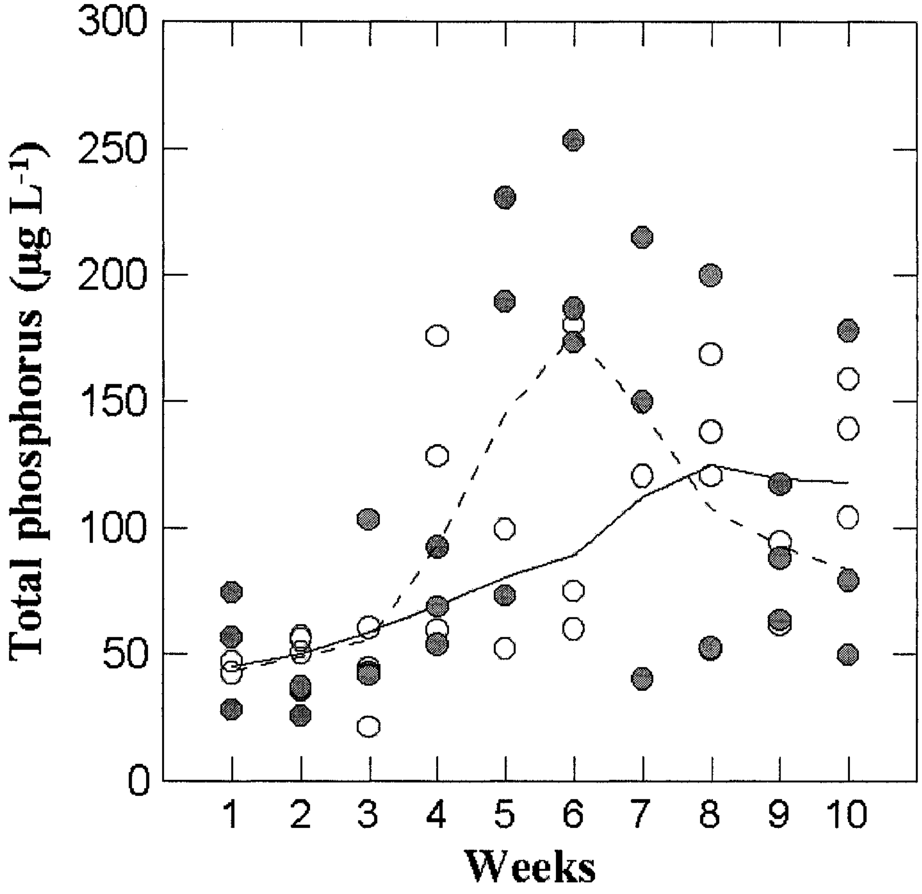


Figure 9. Weekly total phosphorus, from May 14 to July 13, in the tadpoles' habitat of the natural wetland (• and break lo(w)ess) and of the permanent managed wetland (○ and plain lo(w)ess). Smoothing parameter: 0.5.

Total nitrogen : total phosphorus ratio maintained generally between 10 and 20 in both sites for all the monitoring period (Fig. 10). However, values from the natural site was averaged lower (mean 12.6; SD: 4.7) than those from the managed site (mean 16.4; SD: 6.5).

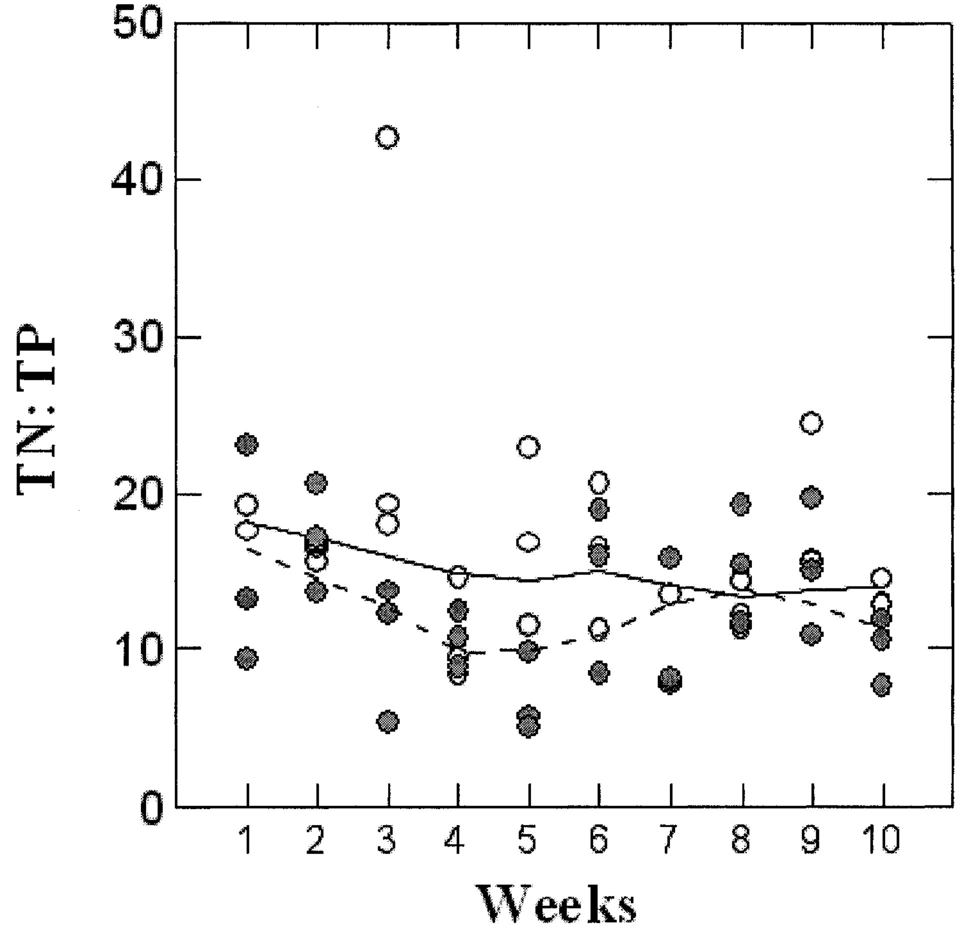


Figure 10. Weekly total nitrogen: total phosphorus ratio, from May 14 to July 13, in the tadpoles' habitat of the natural wetland (● and break lo(w)ess) and of the permanent managed wetland (○ and plain lo(w)ess). Smoothing parameter: 0.5.

During weeks 1 to 6 (May 14 to June 15), sestonic Chl *a* biomass (photosynthetic organisms) was generally low in both the natural and managed wetlands, varying between 0.383 and up to 10.703 µg liter^{-1} (Fig. 11). After the sixth week, sestonic Chl *a* increased drastically (by a factor of five) in the permanent basin, with values up to 26.055 µg liter^{-1} (maximum value in the natural site was 11.426 µg liter^{-1}).

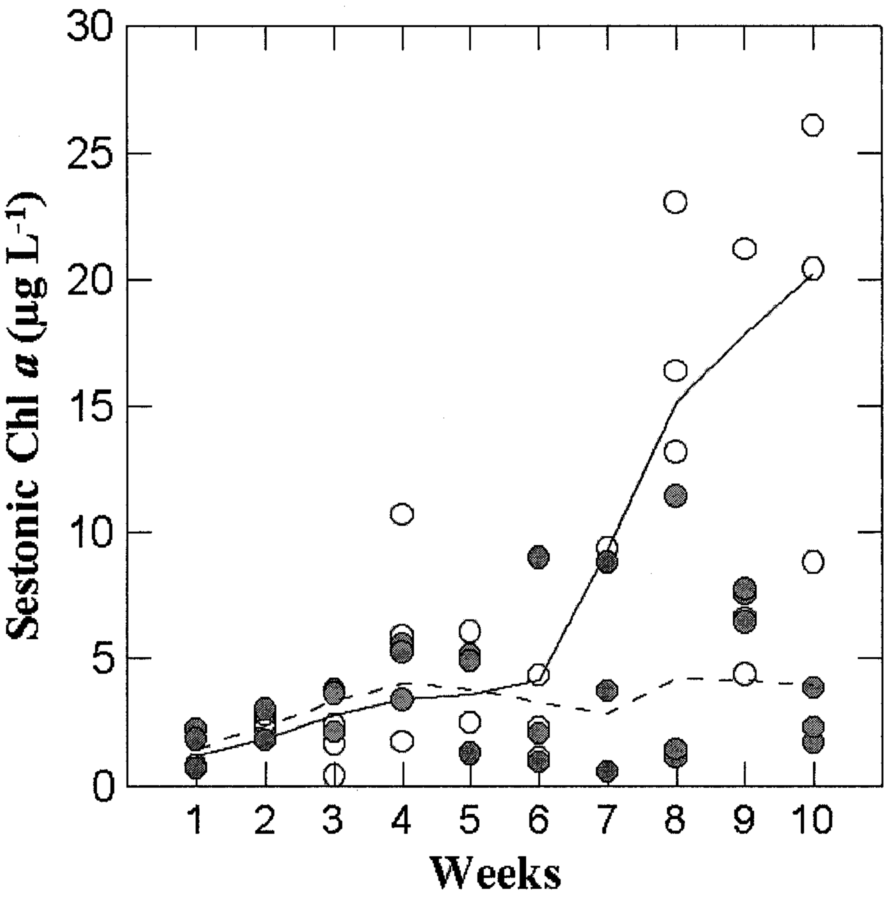


Figure 11. Weekly available sestonic Chl a, from May 14 to July 13, in the tadpoles' habitat of the natural wetland (● and break lo(w)ess) and of the permanent managed wetland (○ and plain lo(w)ess). Smoother tension: 0.5

Sestonic organic matter was equivalent in our two sites during tadpoles' development and increased over time in both sites (Fig. 12). Major augmentation (identified by steeper *lo(w)ess* slope) occurred between weeks 5 and 6 (June 8 to 15) in the natural site and one week later (between weeks 6 and 7: June 15 to 22) in the managed site (Fig. 12).

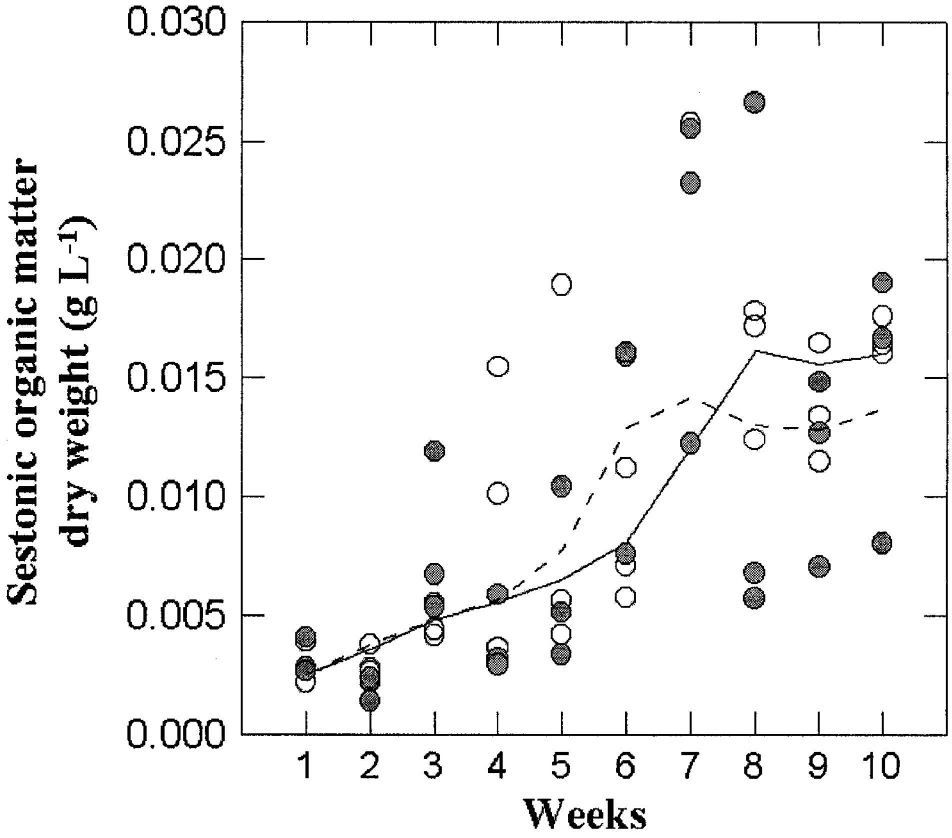


Figure 12. Weekly available sestonic organic matter dry weight, from May 14 to July 13, in the tadpoles' habitat of the natural wetland (● and break lo(w)ess) and of the permanent managed wetland (○ and plain lo(w)ess). Smoothing parameter: 0.5.

No data concerning epiphytic periphyton were collected during the first two weeks of development because at that time tadpoles still relied on their yolk reserve for their development and environmental food availability is probably not relevant. Epiphytic food availability, expressed both as Chl *a* (Fig. 13) and organic matter (Fig. 14), sharply decreased during weeks 3 to 5 (May 25 to June 8) in both sites. During the second half of development (week 6 to 10: June 15 to July 13), both epiphytic Chl *a* and organic matter availability index rose slowly in the managed wetland but remained low in the natural site. The difference is weak for Chl *a* where values of both sites overlap (Fig. 13), but is more evident for organic matter particularly during weeks 7 and 8 (June 22 and 29) (Fig. 14).

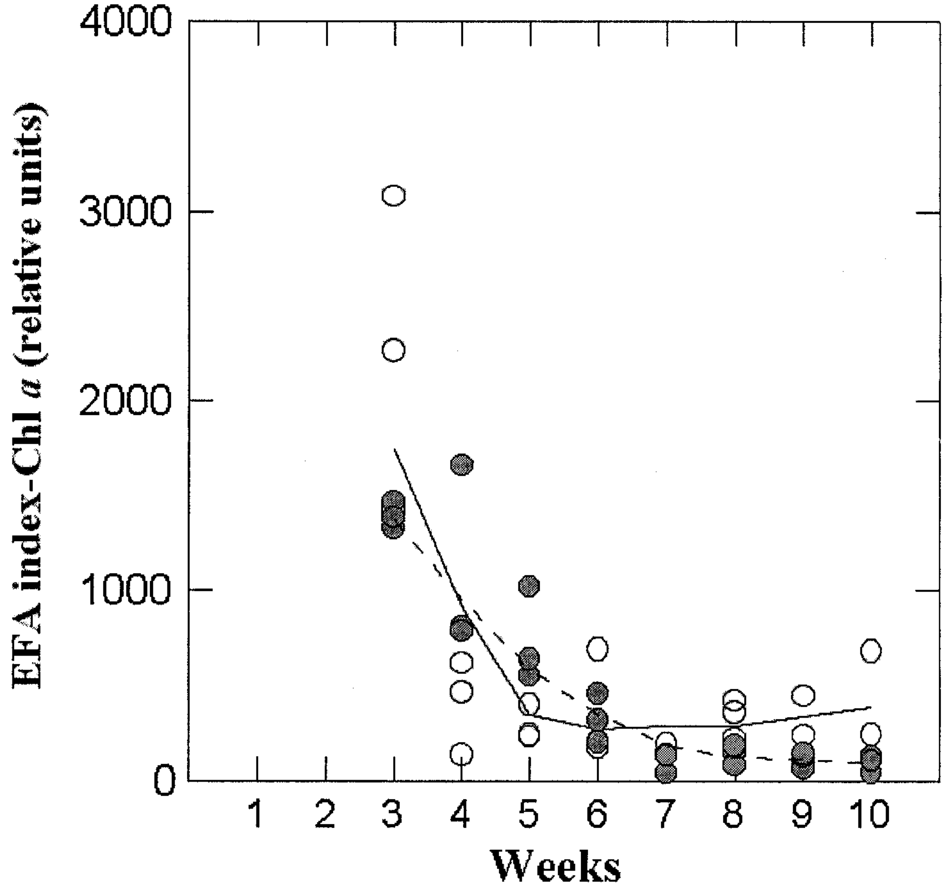


*Figure 13. Weekly epiphytic food availability index for Chl a (calculated as: μg Chl a/cm^2 * Macrophyte percent cover (0-100%) * Water depth (cm)), from May 14 to July 13, in the tadpoles' habitat of the natural wetland (● and break lo(w)ess) and of the permanent managed wetland (○ and plain lo(w)ess). Smoothing parameter: 0.5.*

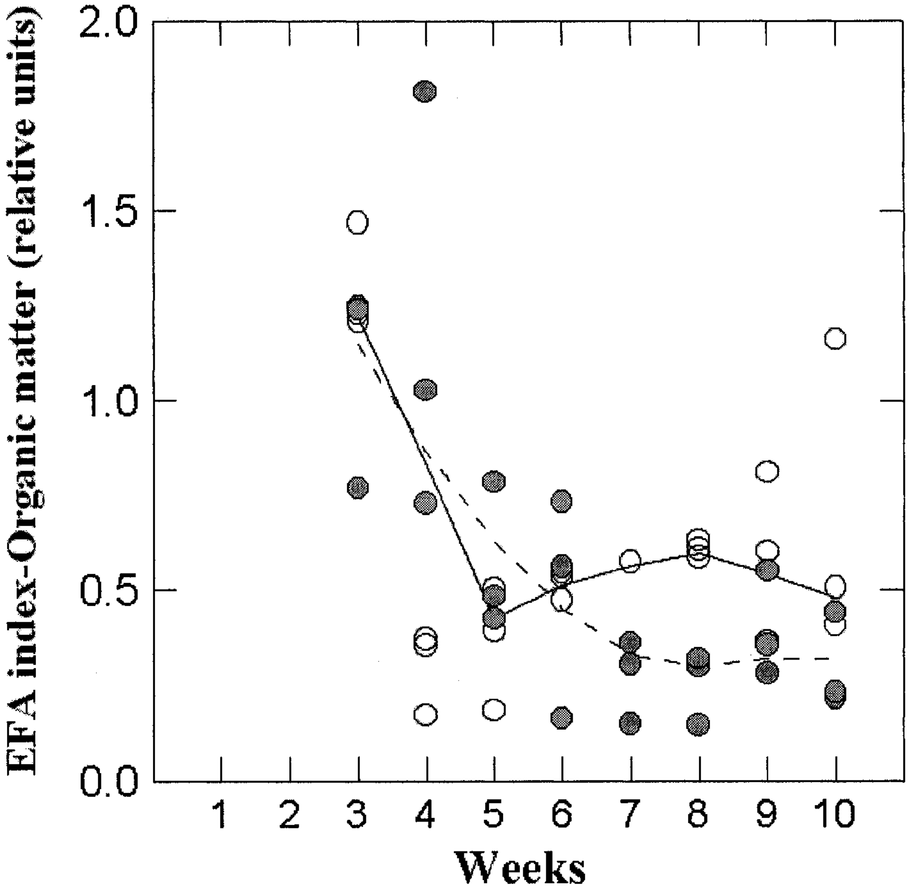


*Figure 14. Weekly epiphytic food availability index for organic matter (calculated as: mg/cm2 organic matter * Macrophyte percent cover (0-100%) * Water depth (cm)), from May 14 to July 13, in the tadpoles' habitat of the natural wetland (● and break lo(w)ess) and of the permanent managed wetland (○ and plain lo(w)ess). Smoothing parameter: 0.5.*

Tadpole density in the managed site was in average 27 % that of the natural site with a mean density of 7 tadpoles/m^3. Tadpole density in the natural site presented two particular episodes of higher density, one at the beginning of the monitoring (week 2: May 18) and one at the end (weeks 8 and 10: June 29 and July 13) (Fig. 15). The first episode corresponds to the aggregation phase of the larvae, which fix themselves with adhesive glands to their egg mass jelly, absorbing yolk until they develop into free-swimming tadpoles and disperse.

This aggregation period was observed in the managed wetland too, but is masked in figure 15 by the extreme values of the natural site. The second episode was only observed in the natural wetland, and corresponds to concentration of tadpoles in pools during low water level events in the St. Lawrence River.

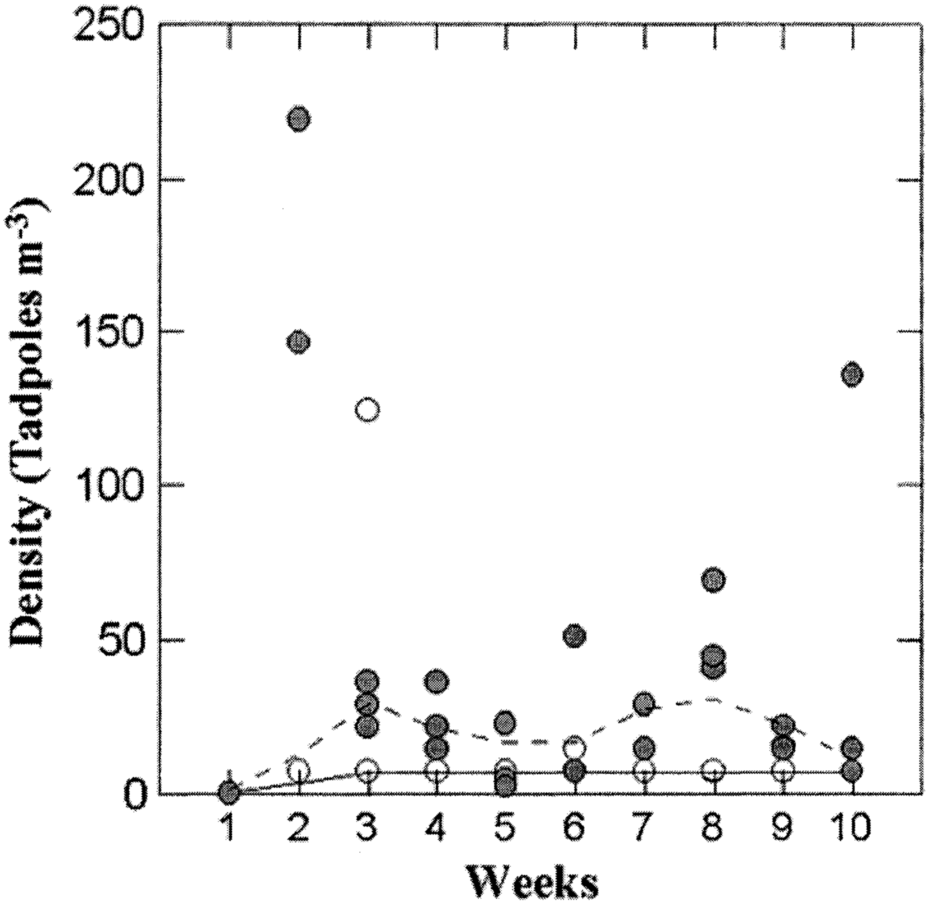


Figure 15. Weekly tadpole density, from May 14 to July 13, in the tadpoles' habitat of the natural wetland (● and break lo(w)ess) and of the permanent managed wetland (○ and plain lo(w)ess). Smoothing parameter: 0.5.

TN:TP ratio, sestonic Chl *a*, tadpoles' density and K_d PAR were entered in a discriminant function which correctly classified the natural and managed stations in 75 to 81% of the cases (Table 1). This discriminant function explained 45% of the total variance.

Plotting of stations on discriminant function scores over time show that tadpoles' environment in the managed wetland was characterized by higher TN:TP ratio, higher sestonic Chl *a* availability, lower tadpole density and lower light extinction coefficient than the tadpoles' natural environment (Fig. 16).

Table 1. Results of discriminant function analysis. The multivariate analysis indicated a significant difference between the two groups of northern leopard frog tadpole's habitats (Wilks' lambda = 0.690, Pillai Trace = 0.310, Lawley-Hotelling trace = 0.450; df= 4, F-ratio= 5.848, P= 0.0006) Eigenvalue: 0.450.

Variables	Canonical discriminant functions	F-to-remove	Tolerance	CDF standardized by within variances
Constant	10.776	.	.	.
TN : TP	2.458	13.42	0.812	0.902
Sestonic Chl a	0.820	9.30	0.744	0.811
Tadpoles' density	-0.350	4.25	0.945	-0.508
Kd PAR	-0.291	0.78	0.884	-0.232

Classification matrix	Natural	Managed	% Correct
Natural	23	7	77
Managed	4	23	85
Total	27	30	81

Jackknifed	Natural	Managed	% Correct
Natural	22	8	73
Managed	6	21	78
Total	28	29	75

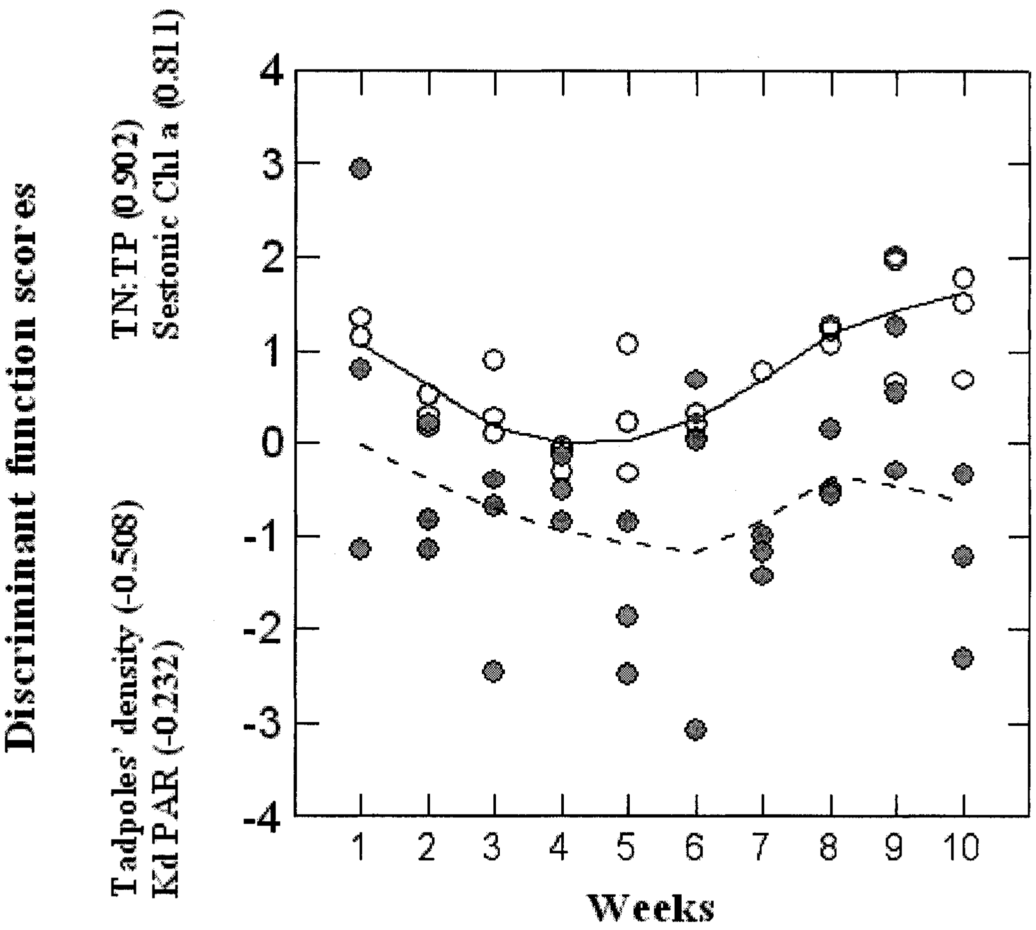


Figure 16. Weekly discriminant function scores, from May 14 to July 13, for tadpoles' habitat in the natural wetland (• and break lo(w)ess) and in the permanent managed wetland (○ and plain lo(w)ess). Smoothing parameter: 0.5.

Development and growth

Figure 17 shows the evolution of the tadpole's development (Gosner stage) in the two sites. The development generally followed the same pattern in the two sites. However, from week 6 to 10 (June 15 to July 13), the intra-variation of Gosner stage is wider in the natural wetland than in the managed one, which indicates longer emergence period for tadpoles in the natural than in the managed wetland.

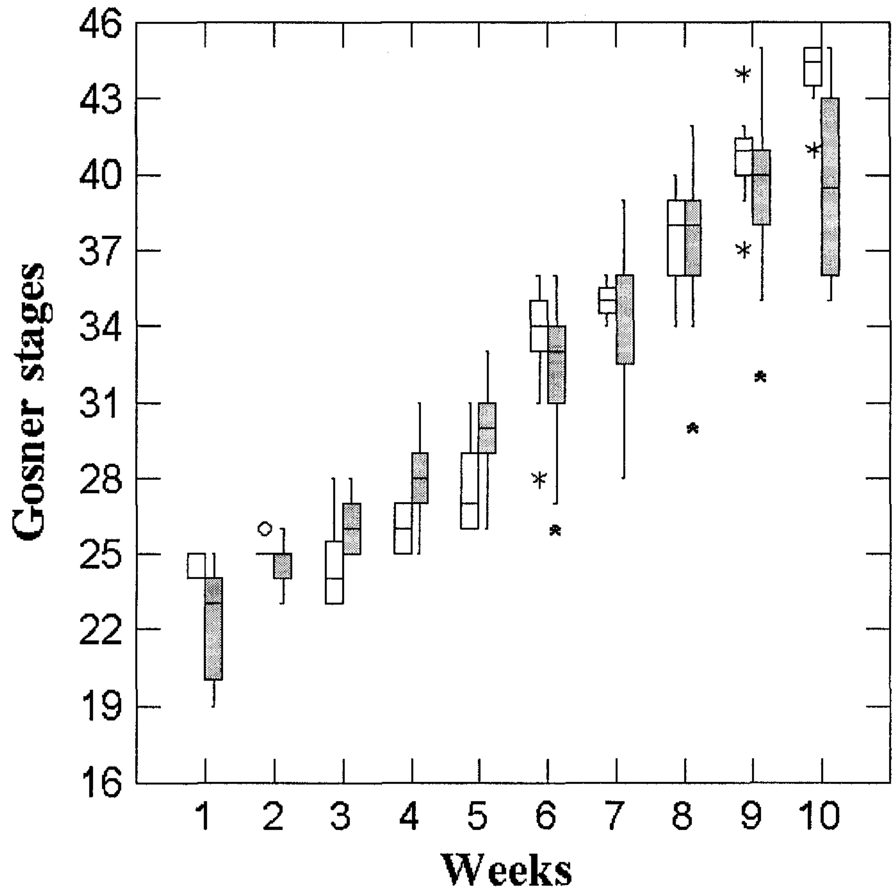


Figure 17. Weekly tadpoles Gosner stages, from May 14 to July 13, in the natural wetland (grey box plot; N: 529) and in the permanent managed wetland (white box plot; N: 233). Asterisks represent values which are 1.5 times the interquartile range (or midrange) and empty circles those which are 3 times this range.

Because Gosner stages are highly correlated with time (Pearson correlation coefficient: 0.941), wet weight of tadpoles were related to Gosner stages instead of week, as in other figures (Fig. 18). This allows comparaison of individual tadpoles' weight for a given Gosner stage. Our results indicate similar growth patterns during the first three to four weeks in both wetlands; afterwards, tadpoles from the managed wetland grew more rapidly than in the natural site between weeks 5 and 9 (June 8 to July 8) (Fig. 18).

The loss of weight observed at week 9 (July 8) in the natural wetland and in week 10 (July 13) in the managed wetland corresponds to the beginning of the metamorphosis in each habitat (Gosner stage 42). At maximum mean wet weight, just before the beginning of the metamorphosis, tadpoles from the managed wetland were more than three times heavier than those from the natural wetland.

Moreover, tadpoles from the natural site attained 100% of their final "week 10" wet weight at week 6 (June 15) and those from the managed wetland attained the same point somewhere between weeks 7 and 8 (between June 22 and 29) (Fig. 19) indicating a longer growth period.

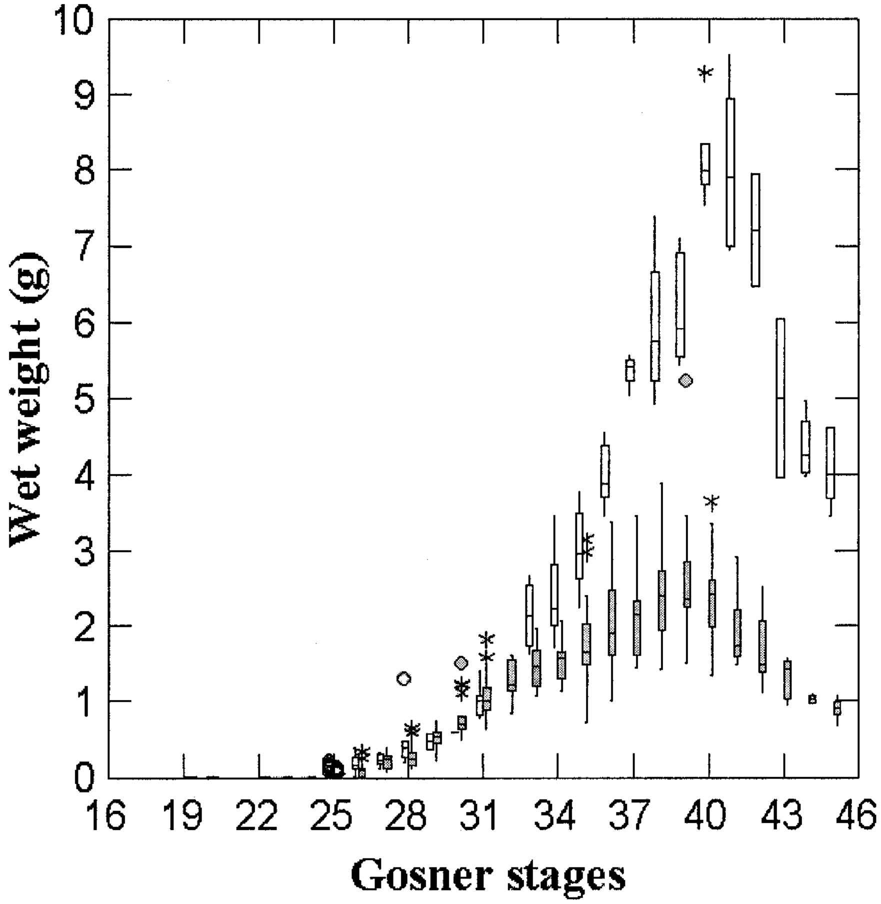


Figure 18. Tadpoles' wet weight by Gosner stage, from May 14 to July 13, in the natural wetland (grey box plot; N: 528) and in the permanent managed wetland (white box plot; N: 233). Asterisks represent values which are 1.5 times the interquartile range (or midrange) and empty circles those which are 3 times this range.

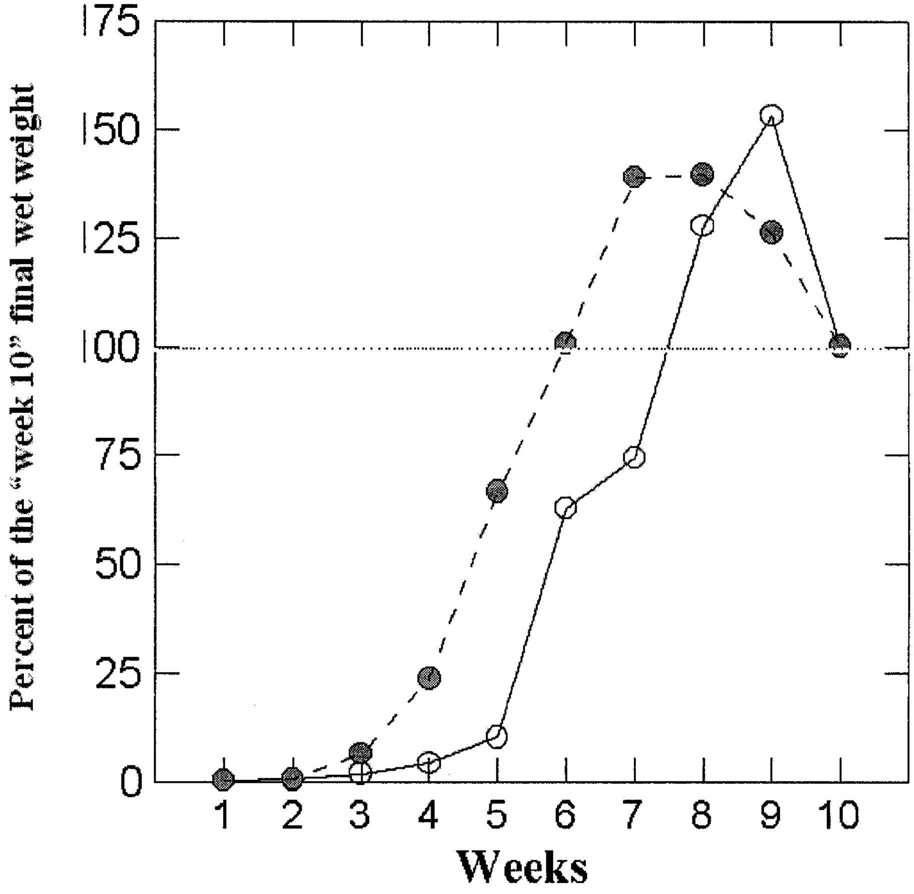


Figure 19. Percent of the "week 10" final wet weight attained by tadpoles in the natural wetland (● and break line) and in the permanent managed wetland (○ and plain line).

Metamorphs

Metamorphs from the managed wetland were significantly heavier than those from the natural site (Natural site: Mean: 1.467g; SD: 0.454; Range; 0.5-3.0; N: 106 and Managed site: Mean: 4.988g; SD: 0.886; Range: 3.0-7.5; N: 89). We were interested to know if this difference in size, between metamorphs from the characterized natural and managed sites, could be generalized to other natural and managed wetlands. The two-tailed t-test on the metamorphs snout-vent length (SVL), showed a highly significant difference ($p < 0.0001$; df: 398) between metamorphs from the permanent managed wetlands, who were longer than those from the natural ones (Fig. 20).

Malformations and predation marks were observed on 2.3% (N: 89) of the metamorphs in the managed site and on 5.7% (N: 106) of the metamorphs in the natural site.

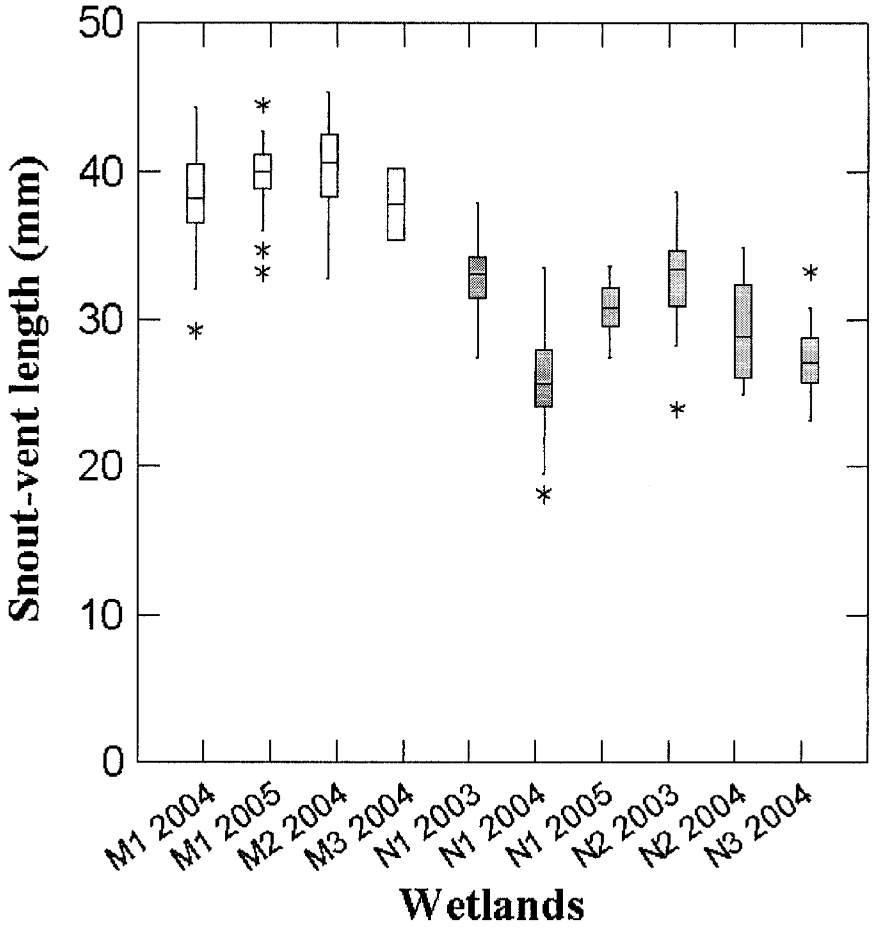


Figure 20. Snout-vent length of metamorphs from three natural bays (grey box plot) and three permanent managed wetlands (white box plot) on the south shore of Lac Saint-Pierre from 2003 to 2005. See Fig.1 to link data to sites. Asterisks represent values which are 1.5 times the interquartile range (or midrange).

Discussion

Permanent managed wetlands as "source habitats" for northern leopard frog

Our first objective was to determine if the northern leopard frog egg masses, laid in permanent managed wetland during early spring, could complete their development without significant abnormalities that could compromise their survival. Based on the interpretation of our results and on the following reflections, we confirm Schueler and Karstad's (2000) hypothesis that NAWMP permanent managed wetlands can be considered as "source habitats" for the northern leopard frog and that those wetlands are therefore probably contributing to the local conservation of this declining species.

We were firstly concerned that the management practices specific to this type of managed wetland, could have profound negative impacts on tadpoles' development success. Maintaining water level by pumping water from the St. Lawrence River could have added to the wetland, a community of tadpole predators. Mud minnow (*Umbra limi*), brown bullhead (*Ictalurus nebulosus*) and invertebrates such as *Odonata* larvae, *Lethocerus*, *Belostoma*, *Notonecta* bugs and parasitic leeches were observed in the permanent managed wetland (Pouliot and Frenette, pers. obs.). Tadpoles' predators were observed in the natural site too, but our unquantifiable impressions suggest that they were less numerous and less diversified (Pouliot and Frenette, pers. obs.). Despite the probable existence of a high predation pressure in the managed site, it seems that a portion of northern leopard frog's tadpoles survived during the 2004 season and that the bulk of egg masses was not just used as food resource by predators. On the other hand, maintaining water level limits the possibility of a "lethal event" in the permanent site. Such "lethal events" exist in other managed wetland conceptions. Temporary flooded managed wetland, designed as migratory stops for waterfowl, cannot support tadpoles to metamorphosis as they are drained at mid-summer (Pouliot and Frenette, pers. obs.). Tadpoles in the permanent managed wetland successfully completed all the normal development steps in the same given wetland, which so could be considered as a "source habitat".

Moreover, tadpoles that developed in the permanent managed wetland grew more rapidly and emerged at a larger size than those from the natural sites. On the basis of the following arguments, we infer that the larger size of the metamorphs in the managed wetland should contribute to increase individual survival which in turn improves the capacity of permanent managed wetlands to act as “source habitats”.

Van Buskirk and Saxer (2001) as also John-Alder and Morin (1990) obtained a positive relationship between metamorphs’ total length and jumping ability, where longer frogs/toad have greater jumping capacities. Considering those observations, combined with the fact that jumping is involved in the escape behavior of northern leopard frog metamorphs (Heinen and Hammond 1997), we hypothesize that longer metamorphs, such as those produced in permanent managed wetlands, should have a better chance to survive to predators as they express anti-predator behaviour more efficiently than smaller ones. This question should be further explored in restraint and semi-natural environments, with metamorphs exposed to a diversity of predators. Metamorphs could be considered as predators too, preying on invertebrates without specific selection and consuming them mainly on their relative abundance (Whitaker 1961). In the context of the optimal foraging theory (OFT), time (energy) invested in searching and manipulating prey is a major factor which affects foraging efficiency (MacArthur and Pianka 1966). Metamorphs’ snout-vent length and mouth width are positively correlated in northern leopard frog metamorphs (Pouliot, unpublished data). We hypothesize that in the context of the OFT, metamorph’s bigger size (and wider mouth) give access to a larger range of prey, which should reduce searching time and enhance access to larger, energy richer, prey. Those elements should increase the cost: benefit ratio and thus enhance the survival of bigger metamorphs. Finally, another advantage linked to a bigger size at metamorphosis is the increased physiological ability to resist to desiccation. Body size will affect this ability as a lower surface: volume ratio (obtained by a larger body) will minimize water loss (Eckert *et al.* 1999) and should enhance resistance to desiccation and survival.

Considering those ecological and physiological advantages conferred by a bigger size, we think that the metamorphs produced by the NAWMP permanent managed wetland don't have less chance to survive than those produced by the natural wetlands, and that managed wetlands can therefore be considered as "source habitats".

Moreover, young leopard frogs are mobile individuals moving as far as 800 and 1000 meters during the first few days of their terrestrial life (Seburn *et al.* 1997; Dole 1971). We thus speculate that the higher survival chance, conferred by larger size, could allow for a higher success during terrestrial movements. This success can promote the colonisation of adjacent good quality habitats that can enhance the capacity of permanent managed wetlands to play a role of "source habitat" in a fragmented landscape. The permanent managed wetlands probably support *in situ* populations but probably also maintain a "frog flow" to other wetlands in the vicinity. This integration of the permanent managed wetlands in the landscape is thus of particular interest in a metapopulation context, as many amphibian populations depend on nearby populations to assure, by individual exchanges, their survival over years (Sjögren 1994; Gill 1978).

Environmental conditions

Our second objective was to describe and compare the environment of the tadpoles in our two wetlands. The environmental similarities between the two sites during the first weeks of the monitoring could be explained by the availability of similar habitats, that is to say the arboresent/shrubby marshes.

With time, tadpoles that develop in the natural site are limited to the marsh, an opened habitat, where they are concentrated in a reduced volume of water. The tadpoles of the managed wetland remain in the shrubby marsh until the metamorphosis.

Most of the variables measured present similar values and seasonal changes. Water depth, water temperature, sestonic organic matter and periphytic food availability were not retained in the discriminant function and are so considered to be relatively similar over time in both sites.

Discriminant analysis groups together (1) TN : TP ratio, (2) K_d PAR coefficient, (3) sestonic Chl *a* and (4) tadpoles density in a discriminant function which associated "tadpoles' habitat" stations to the right wetland (natural or managed) in 75 to 81% of the cases. This function explained 45% of the total variance, as expressed by the eigenvalue. We so considered that the four variables included in the discriminant function could explain an interesting portion of the difference in tadpoles' growth and size at metamorphosis between the managed and the natural wetland.

TN:TP ratio and Kd PAR are known to influence phytoplankton / periphyton community structure (Scheffer *et al.* 1997; Smith 1986). Algae/protista that composed such communities, as diatoms, dinoflagellates, chlorophytes and cyanobacteria are known to be of unequal food quality for herbivores. Diatoms are generally considered as high quality food (Huggins et al. 2004; Bold and Wynne 1985) and cyanobacteria as low quality food (Brett and Muller-Navara 1997). Combine to high K_d PAR coefficient (low light penetration), low (< 29) TN:TP ratio favor cyanobacteria dominance in phytoplanktonic community and so decreased food quality for consumers (Scheffer *et al.* 1997; Smith 1986).

Tadpoles raised in environments with food of higher quality grew more and metamorphosed at bigger size (Kupferberg 1997). Despite that both wetlands had mean TN : TP ratio lower than 29, the natural wetland ratio values were always lower than those from the managed site. Moreover, natural wetland present higher light extinction coefficient most of the time. Considering those characteristics of the natural wetland, we suggest that the available food resource was of lower quality, which could have contributed to the difference in growth and size at metamorphosis. Further investigations should consider direct algae count to establish phytoplankton / periphyton community structure and confirm this hypothesis.

The relations of tadpoles' growth and size at metamorphosis with food quantity have been previously studied. Murray (1990) and Johnson (1991) observed that tadpoles that developed in food rich environments grew more and metamorphosed at bigger size. The notion of food quantity is intuitively related to tadpoles' density as a given quantity of food doesn't represent the same amount for few or many tadpoles to feed on. It's generally accepted that high density results in a slower grow rate and smaller size at metamorphosis (Smith 1983; Wilbur 1977; John and Fenster 1975; Gromko *et al.* 1973).

This has been explained by the competition for available food resources (Johnson 1991; Murray 1990; Berven and Chadra 1988; Wilbur 1977b) and by the high frequency of physical contacts between tadpoles, which is associated with reduced feeding time and growth (John and Fenster 1975; Gromko *et al.* 1973). Our results are consistent with those studies, as smaller size at metamorphosis was observed in the natural site where lower quantity of food and higher densities of tadpoles were observed.

Higher density results also in a greater range between lower and higher Gosner stages in a given cohort, as bigger tadpoles express competitive dominant behavior on smaller ones (Wilbur 1973). Our results suggest such competition due to high density and low sestonic food level, as great Gosner stage ranges were observed only in the natural site, mainly from weeks seven to ten, when density of tadpoles is globally increasing in the natural site but remains constant in the managed one.

The dissimilarities between four environmental variables contributed to our comprehension of the observed size differences in tadpoles and metamorphs from the natural and the managed wetlands. Despite those suggestive results, we proposed that the confinement of tadpoles in pools during the last weeks in the natural wetland, which is not demonstrated by the water depth variable but observed, is probably the proximal cause of environmental changes and therefore, the proximal cause of the size difference.

Water level influences the connectivity between given aquatic habitat areas: higher the water level, greater the connectivity and the area of available habitat for aquatic organisms. In the context of high connectivity, tadpoles can disperse on a greater surface area and forage in lower density, efficiently feeding on available food resources. This scenario prevails during the first weeks of development in both sites. It's during this time, between weeks three and six that tadpoles of the natural site essentially grew, as they attained their metamorphic weight at week six. In the context of low connectivity, available habitat area becomes limiting and tadpoles are forced to aggregate in remnant pools. This drastic change in the tadpoles' habitat of the natural wetland is generated by the falling Saint-Lawrence River water level that begins between week 5 and 6. The water volume reduction concentrated tadpoles and particles in pools. The concentrations of particles is clearly demonstrated by the sudden augmentation of K_d PAR coefficient, TN and TP values between weeks 5 and 6.

Those new environmental conditions should curtail tadpoles' growth for the reasons previously discussed. All those "stressful" conditions should favour the augmentation of development rate over growth, to escape poor conditions, which ultimately results in smaller metamorphs as proposed by Wilbur and Collins (1973).

This hypothesis should be further study as the Saint-Lawrence River water levels have shown great interannual variation over the last decades (Hudon 1997) and such hydrodynamic changes could have profound impact on northern leopard frog' tadpoles development, growth, habitat availability and local survival of small or isolated populations.

Conclusion

In conclusion, we have demonstrated that egg masses laid by northern leopard frogs during the spring can complete their development normally in permanent managed wetlands and these can therefore be considered as "source habitats" for this species. Moreover, the existing potential of those managed wetlands to act as a "frog" provider to neighbor populations is of great interest. Not only in the context of the lake Saint-Pierre floodplain where wetlands are relatively abundant and close one by another, but also in others kinds of ecosystems. In the context of the Boreal forest, like in the James Bay region, where the species is declining and limited to very few breeding sites, the annual "frog flow" to surrounding small populations could be a vital component of the northern leopard frog local survival. The study and the protection of the migratory routes between the managed wetlands and their natural counterparts will be of prime concern for the conservation of the species in such context.

Despite that it's too early to infer that our results could be generalized to permanent managed wetlands elsewhere, it seems to us that if the same management practices are applied to this particular kind of wetland conception, northern leopard frogs should be able to take advantage of the generated environmental conditions and be able to reproduce no matter if the managed wetlands are located in Alberta, Canada or in Iowa, United States.

Future studies should address the effects of the management strategies on the reproduction of other anuran species. Chronology of water pumping, sooner or later during the summer, could probably have an impact on the anuran community composition of the permanent managed wetlands. Species such as the bullfrog (*Rana catesbeiana*), the green frog (*Rana clamitans*) and the mink frog (*Rana septentrionalis*), which breed later in the summer in temperate regions (Cook 1984), could take advantage of a higher water level if it was present during breeding time. For now, it seems that only the leopard frog can breed in those wetlands and we are concerned about the change in the local anuran community structure if only leopard frogs take advantage of those wetlands. Moreover, in western United States, where the bullfrog was introduced and considered as a pest, management practices should be considered to offer a compromise between a positive impact on endangered species and a negative impact on the introduced ones. More knowledge is needed to develop the appropriate management strategies but late season draw-down after that *Rana pipiens* have transformed could be an example of efficient management practices.

The northern leopard frog success story should be considered as an example of the great potential of the NAWMP permanent basin for the conservation of amphibians. Understanding this success was a first step in amphibian conservation using a tool that is already in the conservation tool box, that has a continental distribution and finally, in which the water level can be managed (which indirectly allows for the temporary control of the environmental conditions). The presence of a NAWMP permanent basin in a given area should readily enhance the local success of conservation practices such as the reintroduction of individual adults or the release of young frogs artificially raise from tadpole stages. More than just justifying the great amount of human and financial ressources necessary to annually complete such practices, the presence of a NAWMP permanent basin should permit to gradually withdraw those practices as a self sustainable population establish and shined in vicinity.

Acknowledgements

Special thanks to Jean-François Lapierre, Marianne Lefebvre, Dominique Deshaies, Pascal Lavallé for their help during the 2004 season. Thanks to Jean-François Desroches and Frederick W. Schueler for useful comments on the preliminary version of the text. This research was funded by a) Natural Sciences Research Council of Canada (NSERC) and the Fonds Québécois de la Recherche sur la Nature et les technologies (FQRNT) to J.-J. Frenette and b) FQRNT postgraduate fellowship to D. Pouliot. Work was done according to the S.E.G. permit obtained from the Ministère des resources naturelles et de la faune du Québec. This study is a contribution of the Groupe de Recherche Interuniversitaire en Limnologie (GRIL).

Literature Cited

Batt, B. D. J., M. G. Anderson, C. D. Anderson, and F. D. Caswell. 1989. The use of Prairie Potholes by North American ducks. Pages 204-227. in Van Der Valk, A. editor. Northern Prairie wetlands. Iowa State University Press, Ames, Iowa.

Berven, K. A., and B. G. Chadra. 1988. The relationship among egg size, density and food level on larval development in the wood frog (*Rana sylvatica*). Oecologia 75:67-72.

H.C., Bold, and M.J. Wynne. Introduction to the Algae: Structure and Reproduction. 2nd ed. Prentice-Hall, Inc., Englewood Cliffs, NJ. 720 pp.

Brett, M. T., and D.C. Müller-Navarra. 1997. The role of essential fatty acids in aquatic food web processes. Freshwater Biology 38: 483-499.

Cook, F. R. 1984. Introduction aux amphibians et reptiles du Canada. Mus. Nat. Sci. nat., Musées nationaux du Canada, Ottawa. 211 pages.

COSEPAC. 2002. Évaluation et Rapport de situation du COSEPAC sur la grenouille léopard (*Rana pipiens*) (population des montagnes du Sud et populations boréales de l'Ouest/des Prairies) au Canada. Ottawa. Comité sur la situation des espèces en peril au Canada. Ottawa. Pages 1-46. http://www.cosewic.gc.ca/

Desroches, J.-F., F. W. Schueler, I. Picard, and L.-P. Gagnon. (in press). A herpetological survey of the James Bay area of Québec and Ontario. Submitted to Canadian Field-Naturalist.

Dole, J. W. 1971. Dispersal of recently metamorphosed leopard frogs *Rana pipiens*. Copeia 2:221-228.

D.U.C. (Ducks Unlimited Canada), and NOVE environnement inc. 1990. Projet d'aménagement de la Commune de la Baie-du-Febvre. Étude d'impact sur l'environnement déposée au près du Ministère de l'Environnement du Québec. 134 pages.

Eckert, R., D., Randall, W. Burggren, and K. French. 1997. Animal physiology: mechanisms and adaptations. Fourth edition. W.H. Freeman and Company. New-York, New-York. 723 pages.

FAPAQ (Société de la Faune et des Parcs du Québec). 2003. Le lac Saint-Pierre: Portrait des aménagements et autres sites d'intérêt faunique. Direction de l'aménagement de la faune Mauricie - Centre-du-Québec, Trois-Rivières, Canada. 176 pages

Fisher, S. J., M. R. Anderson, and D. W. Willis. 1998. Total length reduction in preserved yellow perch larvae. North american journal of fisheries management 18:739-742.

Gibbs, E. L., G. W. Nace, and M. B. Emmons. 1971. The live frog is almost dead. Bioscience 21:1027-1034.

Gilbert, M., R. Leclair Jr., and R. Fortin. 1994. Reproduction of the northern leopard frog (*Rana pipiens*) in floodplain habitat in the Richelieu river, P. Quebec, Canada. Journal of Herpetology 28:465-470.

Gill, D. E. 1978. The metapopulation ecology of the red-spotted newt, *Notophtalmus viridescens* (Rafinesque). Ecological Monographs 48:145-166.

Gosner, K. L. 1960. A simplified table for staging anuran embryos and larvae with notes on identification. Herpetologica 16:183-190.

Gromko, M. H., F. S. Mason, and S. J. Smith-Gill. 1973. Analysis of the crowding effect in *Rana pipiens* tadpoles. Journal of experimental zoology 186:63-72.

Heinen, J. T., and G. Hammond. 1997. Antipredator behaviours of newly metamorphosed green frog (*Rana clamitans*) and leopard frogs (*R. pipiens*) in encounters with eastern garter snake (*Thamnophis sirtalis*). American Midland Naturalist 137:136-144.

Huggins, K., J.-J. Frenette, and M. T. Arts. 2004. Nutritional quality of biofilms with respect to light regime in Lake Saint-Pierre (Québec, Canada). Freshwater Biology 49: 945–959.

John, K. R., and D. Fenster. 1975. The effects of partitions on the growth rates of crowded *Rana pipiens* tadpoles. American Naturalist 93:123-130.

John-Alder, H. B., and P. J. Morin. 1990. Effects of larval density on jumping ability and stamina in newly metamorphosed *Bufo woodhousii fowleri*. Copeia 3:856-860.

Johnson, L. M. 1991. Growth and development of larval northern cricket frogs (*Acris crepitans*) in relation to phytoplankton abundance. Freshwater biology 25:51-59.

Hudon, C. 1997. Impact of water level fluctuations on St. Lawrence River aquatic vegetation. Can. J. Fish. Aquat. Sci. 54: 2853-2865.

Kupferberg, S. 1997. The role of larval diet in anuran metamorphosis. Amercian Zoologist. 37: 146-158.

Locky, D. A., J. C. Davies, and B.G. Warner. 2005. Effects of wetland creation on breeding season bird use in boreal eastern Ontario. Canadian Field-Naturalist 119: 64-75.

MacArthur, R. H., and E. Pianka. 1966. On optimal use of a patchy environment. The American Naturalist 100:603-609.

McDiarmid, R. W., and R. Altig. 1999. Research: Materials and Techniques. Pages 7-24. in McDiarmid, R. W., and R. Altig editors. Tadpoles, the biology of anuran larvae. The University of Chicago Press, Chicago, Illinois.

Murray, D. L. 1990. The effects of food and density on growth and metamorphosis in larval wood frogs (*Rana sylvatica*) from central Labrador. Canadian Journal of Zoology 68:1221-1228.

(NAWMP) North American Waterfowl Management Plan, Plan Committee. 2004. North American Waterfowl Management Plan 2004. Strategic guidance: strengthening the biological foundation. Canadian Wildlife Service, U.S. Fish and Wildlife Service, Secretaria de Medio Ambiente y Recursos Naturales. 22 p.

(NAWMP) North American Waterfowl Management Plan, Plan Committee. 1998. North American Waterfowl Management Plan 1998. Strategic guidance: expanding the vision. Canadian Wildlife Service, U.S. Fish and Wildlife Service, Secretaria de Medio Ambiente y Recursos Naturales. 32 p.

Nusch, E. A. 1980. Comparison of different methods for chlorophyll and phaeopigment determination. Arch. Hydrobiologia. Beih. (Ergebn. Limnol.) 14:14-36.

Scheffer, M., S. Rinaldi, A. Gragnani, L. R. Mur, and E. H. Nes. 1997. On the dominance of filamentous cyanobacteria in shallow, turbid lakes. Ecology. 78: 272-282.

Schueler, F. W. and A. Karstad. 2000. Northern Ontario Ducks Unlimited marshes: refugia for leopard frogs? Proceeding of the 5th Canadian amphibian and reptile conservation network annual meeting.

Seburn, C. N. L., D. C. Seburn, and C. A. Paszkowski. 1997. Northern leopard frog (*Rana pipiens*) dispersal in relation to habitat. Pages 64-72. in Green, D. M. editor. Amphibians in decline: Canadian studies of a global problem. Herpetological Conservation, Volume 1. Society for the Study of Amphibians and Reptiles, Saint-Louis, Missouri.

Seburn, C. N. L., and D. C. Seburn. 1998. COSEWIC status report on the northern leopard frog *Rana pipiens* (Southern Mountain and Prairie populations) in Canada, in COSEWIC assessment and status report on the northern leopard frog *Rana pipiens* in Canada. Committee on the Status of Endangered Wildlife in Canada. Ottawa. 1-40 pp.

Smith, D. C. 1983. Factor controlling tadpoles populations of the chorus frog (*Pseudacris triseriata*) on Isle Royale, Michigan. Ecology 64:501-510.

Smith, V. H. 1986. Light and nutrient effects on the relative biomass of blue-green algae in lake phytoplankton. Canadian Journal of Fisheries and Aquatic Sciences. 43:148-153.

Sjögren, P. 1994. Distribution and extinction patterns within a northern metapopulation of the pool frog, *Rana lessonae*. Ecology 75:1357-1367.

Steinwascher, K. 1978. The effect of coprophagy on the growth of *Rana catesbeiana* tadpoles. Copeia 1978:130-134.

Tardif, D., H. Glémet, P. Brodeur, and M. Mingelbier. 2005. RNA/DNA ratio and total length of yellow perch (*Perca flavescens*) in managed and natural wetlands of a large fluvial lake. Canadian journal of fisheries and aquatic sciences. 62:2211-2218.

Van Buskirk, J. and, G. Saxer. 2001. Delayed costs of an induced defense in tadpoles? Morphology, hopping, and development rate at metamorphosis. Evolution 55:821-829.

VanRees-Siewert, K. L., and J.J. Dinsmore. 1996. Influence of wetland age on bird use of restored wetlands in Iowa. Wetlands 16: 577-582.

Wassersug, R. J. 1976. The identification of leopard frog tadpoles. Copeia 2:413-414.

Wetzel, R. G. 2001. Limnology, lake and river ecosystems. Third edition. Academic Press. San Diego, California. 1006 p.

Wetzel, R. G., and. G. E. Likens. 1991. Limnological analyses. Second edition. Springer-Verlag. New-York, New-York. 391 p.

Whitaker, J. O. 1961. Habitat and food of mousetrapped young *Rana pipiens* and *Rana clamitans*. Herpetologica 17:173-179.

Wilbur, H. M. 1977. Density-dependent aspects of growth and metamorphosis in *Bufo americanus*. Ecology 58:196-200.

Wilbur, H. M. 1977b. Interactions of food level and population density in *Rana sylvatica*. Ecology 58:206-209

Wilbur, H. M., and J.P. Collins. 1973. Ecological aspects of amphibian metamorphosis. Science. 182:1305:1314.

CHAPITRE 3

ARTICLE 2

Size of *Rana pipiens* (Northern leopard frog) at metamorphosis after being raised in semi-natural enclosures

Submitted to The Canadian Field-Naturalist

Size of *Rana pipiens* (Northern Leopard Frog) at metamorphosis after being raised in semi-natural enclosures

DANIEL POULIOT, and JEAN-JACQUES FRENETTE

Département de Chimie-Biologie, Université du Québec à Trois-Rivières, 3351 Boul. des Forges, C.P. 500. Trois-Rivières, Québec, Canada. G9A 5H7

Pouliot, D., and J.-J. Frenette. Size of *Rana pipiens* (Northern Leopard Frog) at metamorphosis after been raised in semi-natural enclosures. Canadian Field-Naturalist.

Rana pipiens' tadpoles from two origins: 1- a natural bay of a fluvial lake and 2- a managed wetland developed for waterfowl, have been raised in floating enclosures placed in the managed wetland. Weekly characterization of the enclosures environment didn't showed evident differences. Density, expressed as the survival rate to metamorphosis, shows the stronger difference. The longer and heavier metamorphs emerged from the enclosure with the lower survival rate. Moreover the reduction of tadpoles' density during metamorphosis period is positively related to size at metamorphosis. Our results enhance the importance of density, as an environmental factor, strongly affecting tadpoles during their development and growth.

Des têtards de *Rana pipiens* provenant de deux origines : 1- une baie naturelle d'un lac fluvial et 2- un marais artificiel aménagé pour la sauvagine, ont été élevés dans des enclos flottants placés dans le marais artificiel. La caractérisation hebdomadaire des conditions environnementales dans les enclos n'a pas révélé de différences marquées. La densité, exprimée comme le taux de survie à la métamorphose, était cependant différente d'un enclos à l'autre. Les métamorphes les plus longs et les plus lourds provenaient des enclos ayant présenté les taux de survie les plus faibles. La diminution graduelle de la densité dans les enclos, au fur et à mesure que des têtards se métamorphosaient et quittaient les enclos était positivement associée à la taille à la métamorphose. Nos résultats supportent l'idée que la densité de têtards dans un milieu donné, représente un facteur environnemental de premier ordre, qui affecte grandement les têtards durant le développement et la croissance.

Key Words: *Rana pipiens*, Northern Leopard Frog, density, managed wetlands, enclosures

We studied the development and growth of Northern Leopard Frog tadpoles (*Rana pipiens*) in the Lac Saint-Pierre floodplain (Québec, Canada) (Pouliot and Frenette, in prep). Populations from the natural wetlands located in bays of the fluvial lake where compared to populations from permanent basins, a type of artificial wetland, managed in the context of the North American Waterfowl Management Plan to favour waterfowl populations (details in FAPAQ 2003; NAWMP 2004).

We observed large differences in size at metamorphosis between metamorphs from the natural bays with declining water level and from managed wetlands where water level was kept relatively constant throughout the development. Metamorphs from managed wetlands were about twice the mass of those from the natural sites. Size at metamorphosis is known to be a critical trait in amphibian life history (Wilbur and Collins 1973; Smith 1987) as it affects the individual survival (Altwegg and Reyer 2003) and the reproductive potential of adults (Smith 1987; Semlitsch *et al.* 1988). While this life history trait can be partly controlled genetically (Travis 1981), environmental variables such as density (Gromko *et al.* 1973; John et Fenster 1975; Wilbur 1977), water volume (Crump 1989; Denver *et al.* 1998) and food quantity and quality (Murray 1990; Johnson 1991; Kupferberg 1997) are also known to influence the size at metamorphosis.

In order to test for the contribution of genetic background to the observed difference in size at metamorphosis, which can be drive by the greater survival of larger eggs, we raised tadpoles from both origins in semi-controlled enclosures and exposed to the same environment in a permanent basin. We present our results and discuss possible relationships between environment and growth of both tadpoles origin.

Methods

The study was conducted in the floodplain of Lac Saint-Pierre which is the largest fluvial lake (400 km^2) of the St. Lawrence River. It is characterized by the abundance and diversity of his wetlands. Anurans are well diversified but Northern Leopard Frog (*Rana pipiens*) is the most abundant species (Gilbert *et al.* 1994).

Four *Rana pipiens*' eggs masses, from two breeding sites, were collected on May 05 2005 and brought to the lab. Two of them came from a natural bay (N46° 07', W72° 52') and the two others were from a permanent basin (N46° 08', W72° 47') (Figure 1). Egg masses were raised separately in plastic containers (9L) filled with dechlorinated tap water, at room temperature (16 to 22°C), until hatchling. Water was changed every day and dissolved oxygen was provided with a small aquarium air pump. Following hatchling, 50 larvae (around Gosner stages 21) from each egg mass (total 200 larvae) were randomly chosen and transferred into one of our four experimental enclosures (50 larvae from one given egg mass/enclosure) (Figure 2). Enclosures were installed at the experimental site, the permanent basin (Figure 1), 10 days before the beginning of the experiment to allow growth of algae food resources in the enclosures.

Enclosures were paired: one enclosure containing larvae from the natural wetland ("N" enclosures) was installed side to side to one containing larvae from the managed basin ("M" enclosures). Those two pairs of enclosures were immersed in the water column at locations considered as representative of the "tadpoles' habitat" identified during our previous study at the same site in 2004 (Pouliot and Frenette, in prep). Floating enclosures were made of a wood frame of 122 cm long, 45 cm wide and 22 cm deep. A pocket of about 0.12 m^3, made of mosquito screen (mesh of 1.25mm), was fixed to the wood frame. Density at the beginning of the experiment was 414 tadpoles/m^3, about 2 to 3 times higher than maximal tadpoles' density observed in the wetlands of origin, one year earlier. The Styrofoam floating system maintained the enclosure at the water surface, so that tadpoles were raised within the upper 22 cm of the water column. The open mesh material of the pocket allowed the water and sestonic content to circulate freely between the inside and the outside of the enclosure. At the bottom of the screen pocket, and supported by a wood floor, 24 unpolished sandstone tiles of 225cm^2 (15 x 15cm) were placed to act as substrate for the periphytic algae (Huggins et al. 2004). Periphytic algae grew on the screen pocket too. After larvae were placed in the enclosures, tadpoles were not manipulated. Environment conditions within each enclosure were monitored weekly, during nine weeks.

Water temperature (°C), dissolved oxygen (ppm) and pH were measured with a Hydrolab data sonde (Campbell Scientific) and underwater light regime quantified with a spectroradiometer (Lycor). Sestonic and periphytic food availability, measured as the amount of organic matter (OM) in the water column and fixed to tiles was estimated. Two liters of water from each enclosure were prefiltered in the field on 63 μm to eliminate large zooplanktonic consumers. Water samples were kept in a cooler with ice-packs and filtered the same day on glass-fiber filters (GFF 47μm). Organic matter dry weight (total OM – inorganic OM from burned filters) was measured with a Sartorius balance (0.01mg). For the periphyton, one stripe of 4 by 14 cm was scraped with a sharp razor blade on four randomly chosen tiles (total of 224cm^2, 5% of the total tiles surface). The blended biofilm was mixed in a known volume of distilled water with a cooking mixer. The slurry was filtered the same day on glass-fiber filters (GFF 47 μm) and organic matter dry weight were measure as in the case of sestonic matter but expressed in square centimeter units. At the beginning of the metamorphosis (defined as the first sight of a froglet Gosner stages over 41), enclosures were visited daily. Each day, we collected metamorphosing froglets for examination to the lab. They were kept individually in small Styrofoam container with about half-centimeter of clean water until they completed their metamorphosis (Gosner stage 46). Once metamorphosis completed (always under 72 hours), snout-vent length (mm) and wet weight (mg) were measured with a caliper (Mitutoyo 0.01mm) and a Sartorius balance (0.01mg) respectively. Additional metamorphs from the managed and the natural site were captured with a dip net and weighted (wet weight) for "control" comparison.

Environmental conditions were compared using principal components analysis and discriminant analysis to check for similarities and/or dissimilarities between the two sites. We tested differences in metamorphs' wet weight between enclosures and between origins (Natural bays or Permanent basin) with a non-parametric Kruskal-Wallis one-way analysis of variance. The post-hoc Kolmogorov-Smirnov two-sample test was used to group metamorphs from their weight characteristics.

Non-parametric tests were used because our data did not meet the assumption of homogeneity of variance, essential for parametric ANOVA. Finally, the metamorph's wet weight was plotted against the duration of the larval period for each enclosure to check the potential relationship between the release of the density pressure and the size at metamorphosis.

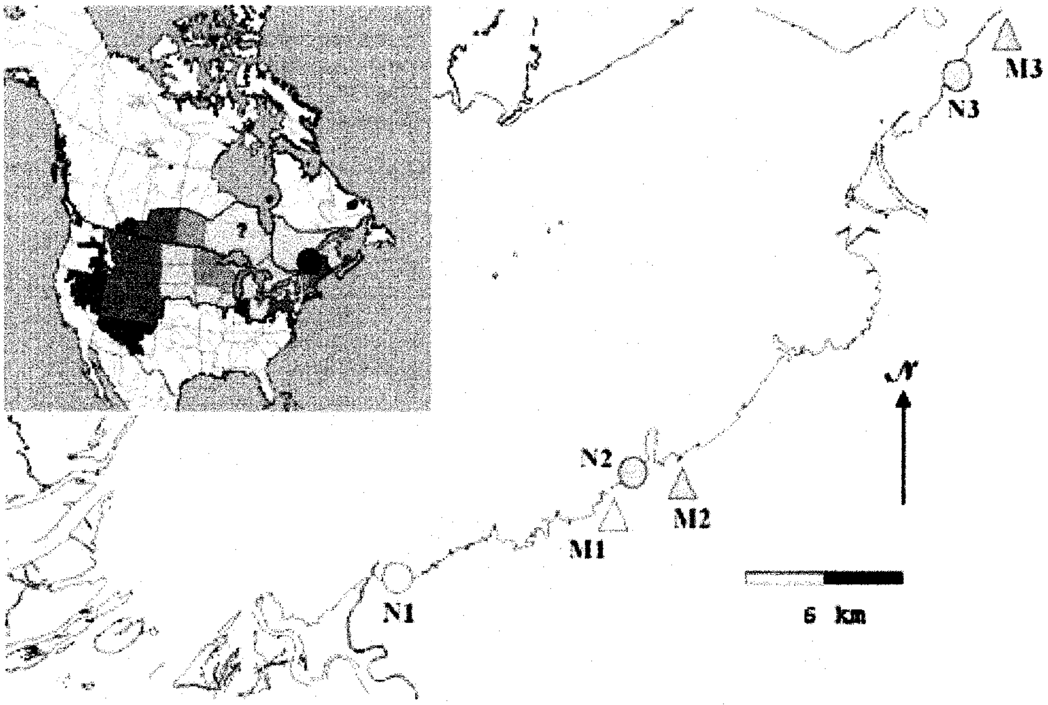


Figure 1. Top-left corner: the circle localized the study area. The Northern Leopard Frog distribution is show in tones of grey following the subnational "S" rank of NatureServe Conservation Stastus; darker the tone more endangered the northern leopard frog is (S5 to S1). The "?" suggest that status in northern Ontario can be more critical than taught, as suggest by Desroches et al. (*in press*). Main image: localisation of the study sites in Lac Saint-Pierre, St. Lawrence River, Canada. N1 and M1 represent locations of natural (N) and managed (M) wetlands where tadpoles' environment, development and growth have been characterized. N2, N3, M2 and M3 refer to stations where only snout-vent length of metamorphs was measured.

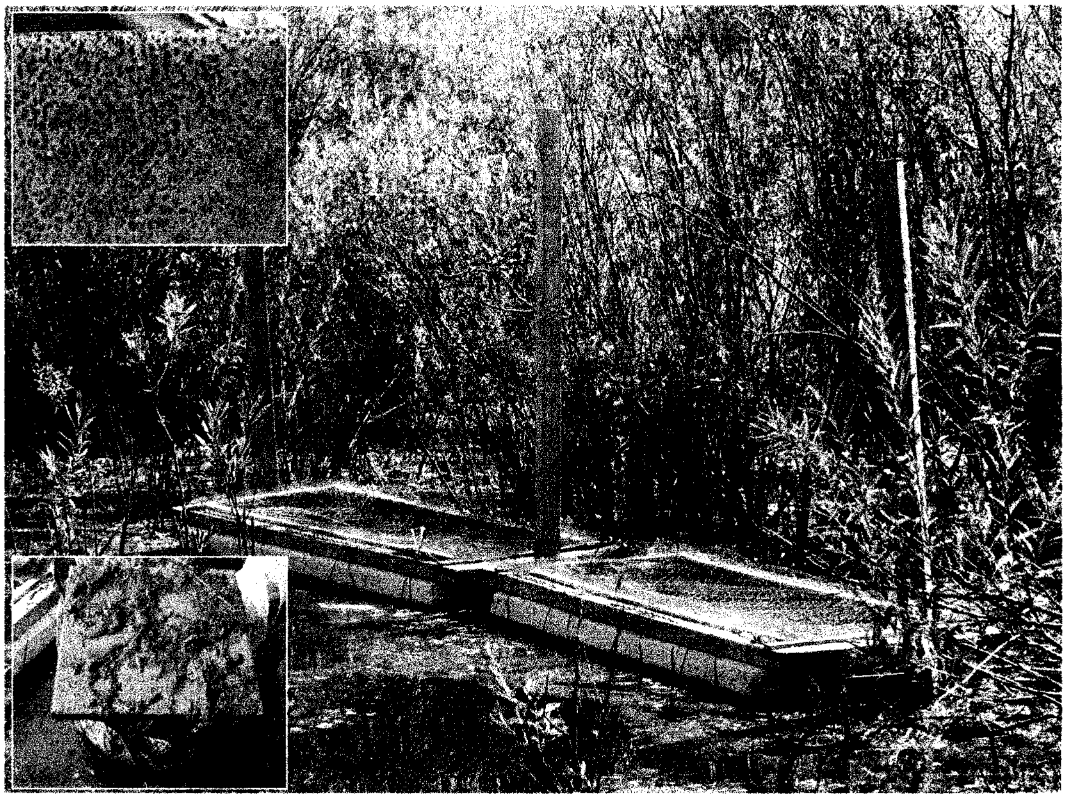


Figure 2. Floating enclosures used to raise tadpoles from two origins in a managed wetland in 2005. Details of tiles partly covered by periphytic algae on top and bottom left corners.

Results

Length of development

From the time where eggs masses were collected in the field on May 05 2005, until the end of the metamorphosis, mean total development length (larval + metamorphosis) was 92 days (n=4; SD: 2,6). Mean metamorphosis period, which corresponds to the time lapse between the observation of the first and the last "leaving" metamorphs was of 30 days (n=4; SD: 3).

Survival rate

Survival rate was highly variable among enclosures (Table 1). Tadpoles from the permanent basin had extreme survival rates, with values of 14 and 66 %, whereas those from the natural site were intermediate (36 and 38 %). Survival rates for each enclosure are summarized in Table 1.

Tadpoles' density

The evolution of the tadpoles' density within enclosures was not monitored to avoid manipulating tadpoles during their development which could increase the risk of injuries and mortalities. However, we could estimate the density just before and during the metamorphosis period in each enclosure, using data from the daily removal of metamorphs. Results indicate that tadpoles' density was unequal in each enclosure during the developmental period as tadpoles' density just prior to metamorphosis was unequal (Table 1). Tadpoles' density in the "natural origin" enclosures was relatively similar but those from the "managed origin" present extreme values (Table 1). Evolution of tadpoles' density during metamorphosis (Figure 3) shows a similar pattern in "natural origin" enclosures: a density drop (a step) at the beginning, as 50% of the froglets emerged within 3 to 6 days after the observation of the first Gosner stage > 41, followed by a much longer period during which individual emergence was sporadic. Enclosure M2, which supported the lowest density at the beginning of the metamorphosis, showed no step, only the prolonged emergence period. Finally, enclosure M1, which had the highest tadpole density at the beginning of the metamorphosis, presented a two-step pattern: a first step where about 25% of the surviving tadpoles emerged, followed by a first plateau, followed by a second step where about 50% of the remaining tadpoles emerged, which is again followed by the second plateau, where individual emergences are spaced out (Figure 3).

Table 1. Comparison of survival rate and tadpoles' density in each of our semi-natural enclosures.

Enclosures	Tadpoles' origin	Survival rate	Density at the beginning of metamorphosis
Enclosure M1	Managed	66 % (33/50)	33 tadpoles ($273/m^3$)
Enclosure M2	Managed	14 % (7/50)	7 tadpoles ($58/m^3$)
Enclosure N1	Natural	36 % (18/50)	18 tadpoles ($149/m^3$)
Enclosure N2	Natural	38 % (19/50)	19 tadpoles ($157/m^3$)

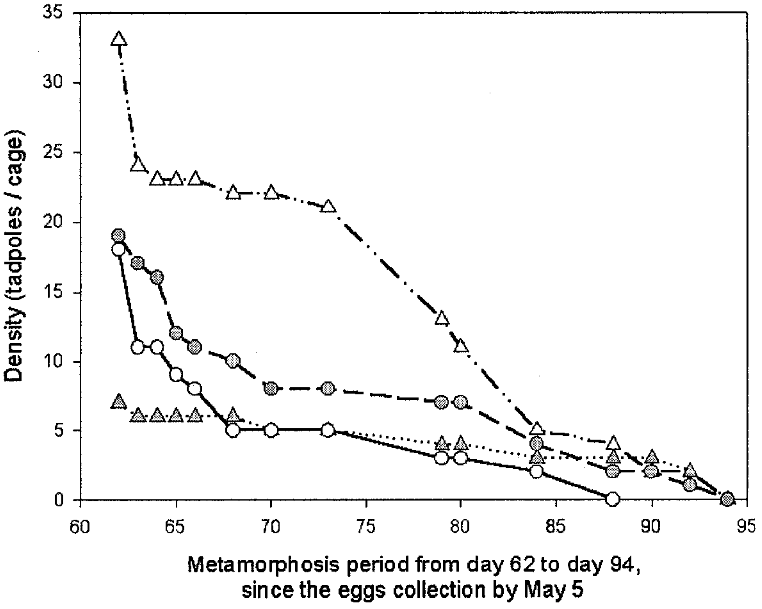


Figure 3. Decreasing tadpoles' density in each of the four enclosures, during the metamorphosis period. Symbols of paired enclosures are of the same color.

(N1: ○, N2: ●, M1: △, M2: ▲).

Metamorphs' wet weight

Non-parametric Kruskal-Wallis one-way analysis of variance on metamorphs' wet weight between enclosures was significant (p=0.013). Metamorphs from enclosure M2 were heavier than those from other enclosures. This difference originates from the bimodal distribution of data in enclosure M2. In fact, half the metamorphs (3/6) from this enclosure were similar in size to those from enclosures M1, N1 and N2 (Table 2). No significant difference was observed in relation to their origin (Natural *vs.* Permanent Basin) (Mann-Whitney U test statistic 749.000 p=0.253). Details of their morphology appear in Table 2.

Table 2. Comparison of metamorphs' size for individuals raised in semi-natural enclosures with those captured in breeding sites of origin (controls). Cells present mean, standard deviation, range and sample size.

Enclosures	Tadpoles' origin	Snout-vent length (mm)	Wet weight (g) of Gosner stage 46
Enclosure M1	Managed	23.92 (SD: 1.78) 20.55-27.08 n: 29	0.97 (SD: 0.25) 0.64-1.63 n: 29
Enclosure N1	Natural	22.81 (SD: 3.74) 18.09-30.16 n: 18	0.98 (SD: 0.55) 0.50-2.13 n: 18
Enclosure M2	Managed	29.07 (SD: 3.96) 23.48-33.61 N: 6	2.22 (SD: 1.22) 0.92-3.44 n: 6
Enclosure N2	Natural	24.63 (SD: 2.61) 21.52-30.69 n: 19	1.11 (SD: 0.42) 0.64-1.92 n:19
Control	Managed	39.53 (SD: 2.72) 33.18-44.46 n: 19	5.948 (SD: 1.49) 3.081-8.272 n: 19
Control	Natural	30.76 (SD: 1.70) 27.38-33.64 n: 31	2.97 (SD: 0.58) 1.83-4.56 n: 31

Kolmogorov-Smirnov two samples test applied to the six categories of metamorphs (enclosures M1, M2, N1, N2 and Controls M and N) classified them into three groups. The first group is composed of the smallest metamorphs from enclosures M1, N1 and N2.

The second group of intermediate size is composed of metamorphs from enclosure M2 and Control N. Finally the heaviest metamorphs were produce in the permanent basin, outside the enclosure and is composed only of Control M metamorphs (Figure 3). Weight at metamorphosis was negatively related to survival rate in the enclosures as seen in Figure 4.

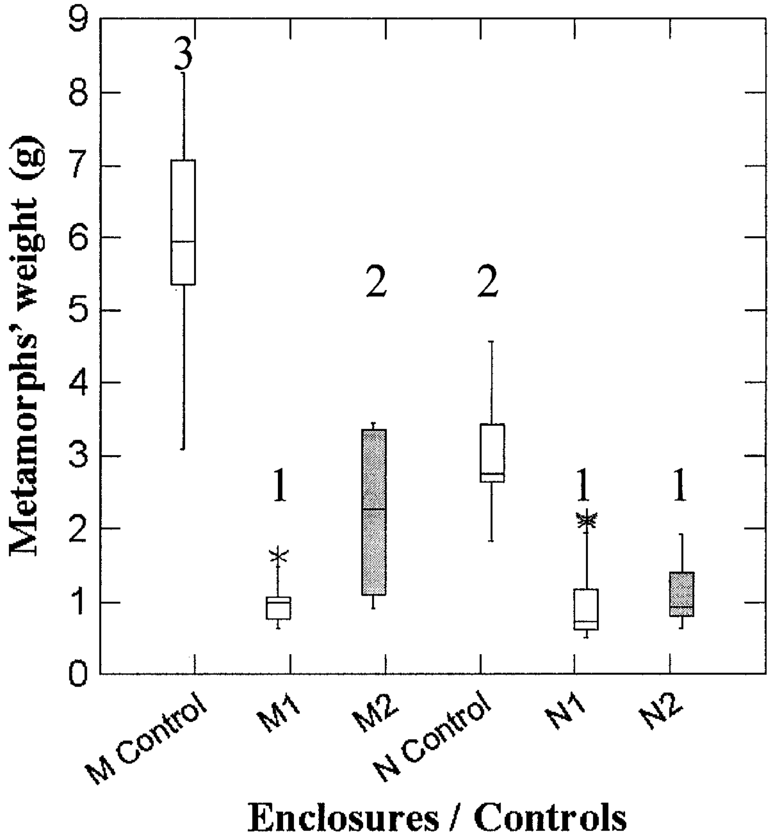


Figure 4. Metamorphs' wet weight for each enclosure and control. Number 1, 2 and 3 refers to the group number; Kolmogorov-Smirnov two samples test was used to classify the metamorphs.

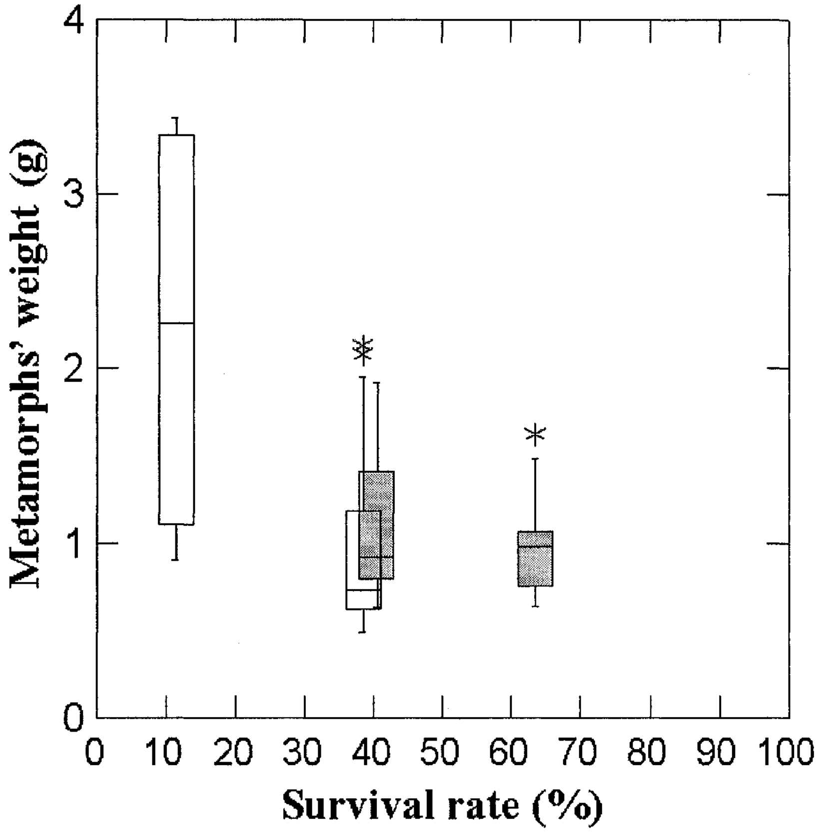


Figure 5. Metamorphs' wet weight in relation to survival rate for tadpoles raised in four semi-natural enclosures. Box plots of the same color represent paired enclosures. From left to right: enclosure M2, N1, N2 and M1.

The general relationship between the weight at metamorphosis and the duration of the development is positive in all our enclosures (Figure 5). In all enclosures, individuals that stay longer in the aquatic medium grow more and metamorphosed at bigger size. While there is a positive relationship in all four enclosures, the slope varied among enclosures. Again, enclosures with tadpoles from the permanent basin, M1 and M2, exhibited contrasting slopes, whereas those with tadpoles from the natural site were intermediate.

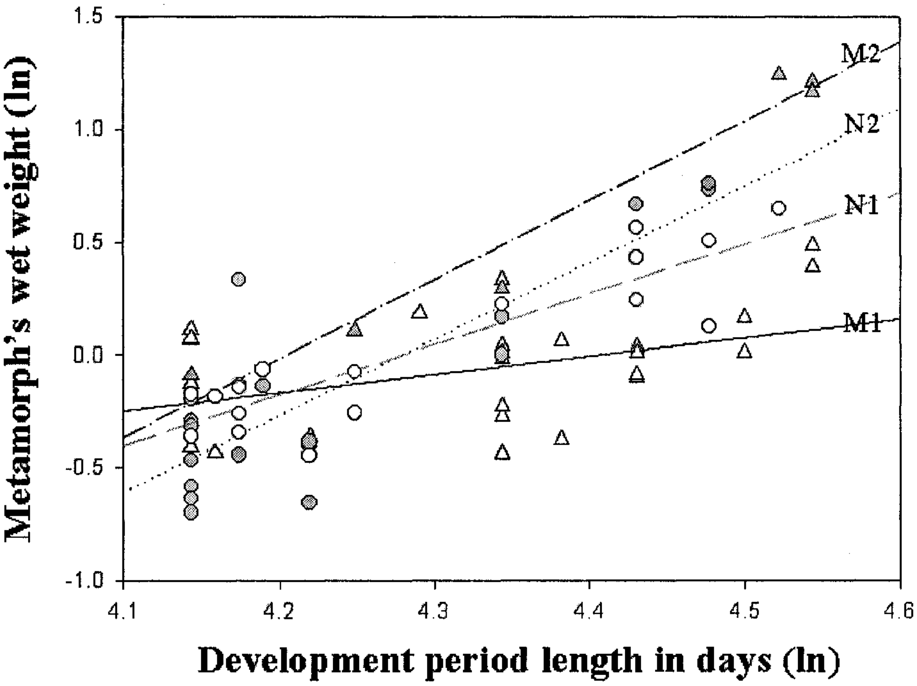


Figure 6. Metamorphs' wet weight in relation to the duration of the total developmental period (larval + metamorphosis) in days. Symbols of paired enclosures are of the same color. (N1: ○, N2: ◍, M1: △, M2: ▲).

Environmental variables

Other environmental variables present less difference than density. Food availability, sestonic and periphytic, was similar between enclosure sites. Multivariate analysis, principal component analysis and discriminant analysis did not reveal any significant pattern between sites suggesting great similarities and weak differences between environmental conditions of each enclosure. Table 3 summarizes results for each variable.

Table 3. Comparison of environmental characteristics in tadpoles' enclosures from a permanent basin managed for waterfowl, Lac Saint-Pierre, St. Lawrence River, Canada. In each case: Mean, Standard Deviation, Range and Sample size.

Variables / Enclosures	Paired A Eggs' origin M Enclosure # M1	Paired B Eggs' origin M Enclosure # M2	Paired A Eggs' origin N Enclosure # N1	Paired B Eggs' origin N Enclosure # N2
Sestonic Organic Matter (g/L)	0.00251 (SD : 0.00060) 0.00155-0.00304 n :9	0.00238 (SD : 0.00065) 0.00149-0.00315 n :9	0.00221 (SD : 0.00049) 0.00151-0.00295 n :9	0.00218 (SD : 0.00051) 0.00176-0.00292 n :9
Periphytic Organic Matter (g/cm^2)	0.782 (SD: 0.668) 0.200-2.123 n: 8	0.915 (SD: 0.947) 0.245-3.127 n: 8	0.516 (SD: 0.239) 0.180-0.869 n: 8	0.964 (SD: 0.327) 0.379-1.387 n: 8
pH	7.09 (SD : 0.32) 6.71-7.49 n : 7	7.01 (SD : 0.38) 6.52-7.45 n : 7	7.08 (SD : 0.30) 6.69-7.58 n : 7	7.04 (SD : 0.47) 6.50-7.55 n : 7
Dissolved O_2 (ppm)	4.45 (SD : 2.40) 1.57-7.79 n : 7	4.05 (SD : 2.53) 1.33-7.70 n : 7	4.48 (SD : 2.20) 1.46-7.57 n : 7	3.92 (SD : 2.70) 0.98-7.99 n : 7
Water Temperature °C	19.97 (SD : 5.12) 11.52-24.53 n : 7	19.77 (SD : 4.78) 11.93-24.13 n : 7	19.95 (SD : 5.01) 11.71-24.44 n : 7	19.77 (SD : 4.79) 11.88-24.28 n : 7

Discussion

Enclosures

Despite that tadpoles are regularly known to be filter feeder (Hendricks 1973), they are also recognized as periphytic scrapers (Hourdry 1993; Altig and McDiarmid 1999). Good enclosures should have provided food resources from both origins: sestonic and periphytic. While periphytic algae can grow on the screen surface, its quantification is quite delicate, as scraping the screen pocket with a sharp razor blade could result in pocket breakage and tadpoles to escape. The use of tiles as substrate for periphytic algae was mainly used by limnologists who were interested in the effect of environmental conditions on alga growth (see Huggins *et al.* 2004; Ranvestel *et al.* 2004). Unglazed tiles can be easily scraped without any fear of breaking the enclosure, they allow quantification of the food availability and they provide a good substrate for periphytic algae which represent a major food supply for tadpoles.

Survival rate

Tadpoles' survival rates observed in this experiment are higher than what is generally reported for natural populations. Survival rates in natural anuran populations have been estimated to 4% (Herreid and Kinney 1966: *Rana sylvatica*; Brockelman 1968 in Calef 1973: *Bufo americanus*), 5% (Calef 1973: *Rana aurora*), 1 to 6% (Hine et al. 1981: *Rana pipiens*) and less than 8.5% (Turner 1962 in Calef 1973: *Rana pretiosa*). Our high survival rates, from 14 to 66%, are probably the result of a generally low predation pressure and low grazing competition as we weekly extracted from the enclosures easily visible predators and grazers such as gastropods.

The lower survival rate of enclosure M2 (7/50 or 14 %) is probably explained by a higher predation pressure which prevailed in that enclosure. Two of the seven metamorphs who survived until metamorphosis lacked respectively, one and two hind legs.

Moreover, we found only a few odonate larvae (about 1 or 2) in all enclosures, except in enclosure M2, where as much as 7 odonate larvae were collected. Odonate larvae are known to eat tadpoles (Calef 1973) and their higher abundance in this particular enclosure probably explains the lower survival rate.

Survival rate in enclosure M1 (66%) is particularly high and difficult to interpret. It's possibly an anecdotal result, as the other "managed" enclosure give the opposite result, the lower survival rate. However, a hypothesis could be that those tadpoles, which originated from the same wetland where they were raised, are more efficient in using available food resource in the managed site than tadpoles from the natural bay (which were the "alien" tadpoles). The possibility of morphological/genetical differences between a "natural form" and a "managed form" of Northern Leopard Frog tadpoles should be further investigated.

Density and size at metamorphosis

The relationship between survival rate and size at metamorphosis is negative. Higher survival rate was related to smaller mean metamorphs. This suggests a trade-off between food/space availability and number/size of individuals produced. Survival rate is in fact another way to express density in our enclosures. A high density is known to affecte anuran tadpoles' development by limiting available food resources thus reducing growth rate (Gromko *et al.* 1973; John and Fenster 1975; Wilbur 1977; Smith 1983). Considering that there is no difference in food availability between enclosures, and so that surviving tadpoles have to share/compete relatively the same amount of available food, the intra-enclosure competition for food can explain our results.

Metamorphosis period

Metamorphosis period can be viewed as an explosive emergence period which lasts for a brief time. But, in 2004, we observed that, in a natural bay of Lac Saint-Pierre tadpoles were confined to available pools in open marsh, as the St. Lawrence River water level was decreasing significantly (Pouliot and Frenette, in prep.).

At that time, only 25 % of the tadpoles were ready to emerge (Pouliot and Frenette in prep.). From this observation we thought that the emergence period is undoubtedly not a short period of time but a relatively long process during which tadpoles' environment continues to change and to influence the development of remnant tadpoles. The results of the present study firstly confirmed that under specific environmental conditions, metamorphosis period could take place on a long period of time, as it happened for about a month in our enclosures. Moreover, they suggest that the reduction of density over time, during the metamorphosis, could promote growth of remnant tadpoles as they emerge at bigger size.

The last three tadpoles to emerge in the enclosure M2 (lowest density) emerged as a second group of metamorphs, as their sibling emerged earlier (see Figure 6). They were much bigger than their siblings and probably took advantage of the low density that occured during most of the metamorphosis period. In higher density context, as in enclosure M1, the emergence pattern was characterized by three cohorts, each displaying bigger metamorphs than the preceding. Following Wilbur and Collins (1973) and based on our results, environmental stress for tadpoles in our enclosures, as competition for food generated by a high density of tadpoles, decreased over time. Remnant tadpoles take advantage of those changing conditions, grow more rapidly and achieve metamorphosis at bigger size.

Conclusion

The raising of tadpoles from two different origins in semi-natural enclosures didn't find a genetical difference between populations in a size difference at metamorphosis. In our case, density as an environmental factor affecting individual growth seems to have determinied the quality of growth between each enclosure and in a given enclosure during the metamorphosis period. However, impressive survival rate in one of our "at home" enclosures, raises the idea of local adaptation to the particular wetland categories (habitat), enhancing the efficiency to use resources. This hypothesis could be further investigated in experimental, controlled, environments.

Acknowledgements

Special thanks to Guillaume Blanchette-Martineau, Patrice Thibeault and Jean-François Lapierre for their help during the 2005 season. Thanks to Jean-François Desroches and Frederick W. Schueler for useful comments on the preliminary version of the text. This research was funded by a) Natural Sciences Research Council of Canada (NSERC) and the Fonds Québécois de la Recherche sur la Nature et les technologies (FQRNT) to J.-J. Frenette and b) FQRNT postgraduate fellowship to D. Pouliot. Work was done according to the S.E.G. permit obtained from the Ministère des resources naturelles et de la faune du Québec. This study is a contribution of the Groupe de Recherche Interuniversitaire en Limnologie (GRIL).

Literature Cited

Altig, R., and R.W. McDiarmid. 1999. Body plan: development and morphology. In R.W. McDiarmid and R. Altig (eds.), Tadpoles: the biology of anuran larvae, pp. 24-51. University of Chicago Press, Chicago.

Altwegg, R., and H.U. Reyer. 2003. Patterns of natural selection on size at metamorphosis in water frogs. Evolution 57: 872-882.

Calef, G.W. 1973. Natural mortality of tadpoles in a population of *Rana aurora*. Ecology 54 : 741-758

Crump, M.L. 1989. Effect of habitat drying on developmental time and size at metamorphosis in *Hyla pseudopuma*. Copeia 1989: 794-797.

Denver, R.J., N. Mirhadi, and M. Phillips. 1998. Adaptative plasticity in amphibian metamorphosis : Response of *Scaphiopus hammondii* tadpoles to habitat dessication. Ecology 79: 1859-1872.

Desroches, J.-F., F. W. Schueler, I. Picard, and L.-P. Gagnon. (in press). A herpetological survey of the James Bay area of Québec and Ontario. Submitted to Canadian Field-Naturalist.

FAPAQ (Société de la Faune et des Parcs du Québec). 2003. Le lac Saint-Pierre: Portrait des aménagements et autres sites d'intérêt faunique. Direction de l'aménagement de la faune Mauricie - Centre-du-Québec, Trois-Rivières, Canada. 176 pages.

Gilbert, M., R. Leclair Jr., and R. Fortin. 1994. Reproduction of the northern leopard frog (*Rana pipiens*) in floodplain habitat in the Richelieu river, P. Quebec, Canada. Journal of Herpetology 28:465-470.

Gosner, K. L. 1960. A simplified table for staging anuran embryos and larvae with notes on identification. Herpetologica 16:183-190.

Gromko, M. H., F. S. Mason, and S. J. Smith-Gill. 1973. Analysis of the crowding effect in *Rana pipiens* tadpoles. Journal of experimental zoology 186:63-72.

Hendricks, F.S. 1973. Intestinal contents of *Rana pipiens* Schreber (Ranidae) larvae. Southwestern Naturalist 18: 99-101.

Herreid II, C.F. and S. Kinney. 1966. Survival of Alaskan woodfrog (Rana sylvatica) larvae. Ecology 47: 1039-1041.

Hine, R.L., B.L. Les and, B.F. Hellmich. 1981. Leopard frog populations and mortality in Wisconsin, 1974-76. Technical Bulletin No. 122. Department of natural resources. Madison, Wisconsin. 38 pages.

Hourdry, J. 1993. Passage to the terrestrial life in amphibians: events accompanying this ecological transition. Zoological science 10:715-731.

Huggins, K., J.-J. Frenette, and M.T. Arts. 2004. Nutritional quality of biofilms with respect to light regime in Lake Saint-Pierre (Québec, Canada). Freshwater Biology 49: 945-959.

John, K. R., and D. Fenster. 1975. The effects of partitions on the growth rates of crowded *Rana pipiens* tadpoles. American Naturalist 93:123-130.

Johnson, L. M. 1991. Growth and development of larval northern cricket frogs (*Acris crepitans*) in relation to phytoplankton abundance. Freshwater biology 25:51-59.

Kupferberg, S. 1997. The role of larval diet in anuran metamorphosis. American Zoologist 37:146-158.

Martin, C., J.-J. Frenette, and J. Morin. 2005. Changes in the spectral and chemical properties of a water mass passing through extensive macrophyte beds in a large fluvial lake (Lake Saint-Pierre, Québec, Canada). Aquatic Sciences 67: 196-209.

Murray, D. L. 1990. The effects of food and density on growth and metamorphosis in larval wood frogs (*Rana sylvatica*) from central Labrador. Canadian Journal of Zoology 68:1221-1228.

(NAWMP) North American Waterfowl Management Plan, Plan Committee. 2004. North American Waterfowl Management Plan 2004. Strategic guidance: strengthening the biological foundation. Canadian Wildlife Service, U.S. Fish and Wildlife Service, Secretaria de Medio Ambiente y Recursos Naturales. 22 p.

Pouliot, D., and **J.-J. Frenette.** In prep. North American Watefowl Management Plan permanent basin as an effective tool for northern leopard frog (*Rana pipiens*) conservation.

Ranvestel, A.W., K.R. Lips, C.M. Pringle, M.R. Whiles, and R.J. Bixby. 2004. Neotropical tadpoles influence stream benthos: evidence for the ecological consequences of declinein amphibian populations. Freshwater Biology 49: 274-285.

Semlitsch, R.D., D. E. Scott, and J.H.K. Pechmann. 1988. Time and size at metamorphosis related to adult fitness in *Ambystoma talpoideum*. Ecology 69:184-192.

Smith, D. C. 1983. Factor controlling tadpoles populations of the chorus frog (*Pseudacris triseriata*) on Isle Royale, Michigan. Ecology 64:501-510.

Smith, D.C. 1987. Adult recruitment in chorus frogs: effects of size and date at metamorphosis. Ecology 68:344-350.

Travis, J. 1981. Control of larval growth variation in a population of *Pseudacris triseriata* (Anura: hylidae). Evolution 35: 423-432.

Wilbur, H. M. 1977. Density-dependent aspects of growth and metamorphosis in *Bufo americanus*. Ecology 58:196-200.

Wilbur, H.M., and J.P. Collins. 1973. *Ecological aspects of amphibian metamorphosis*. Science 182 : 1305-1314.

CHAPITRE 4

ARTICLE 3

Pool boat for the study of tadpoles' habitats

Submitted to Herpetological Review

Pool boat for the study of tadpoles' habitats

Daniel Pouliot and Jean-Jacques Frenette

Université du Québec à Trois-Rivières – Département de chimie-biologie – Laboratoire d'écologie aquatique
e-mail (DP) : daniel.pouliot@uqtr.ca
e-mail (JJF) : jean-jacques.frenette@uqtr.ca

Despite the large amount of information in the literature concerning tadpole ecology, few data are available about the habitat requirements of tadpoles. By skipping those early stages of life, we often miss a great opportunity to better understand the biology of frogs in general and also why frogs are declining in North America. Changed tadpoles' habitat could explain, as one of a great number of causes (Kiesecker et al. 2001), a part of the known decline (Houlahan *et al.* 2000).

One reason that can explain the lack of « *in situ* » data about tadpoles' habitat could be the difficulty of working in the anuran's habitats. More specifically these wetlands are too shallow to access from a boat but at the same time, to deep to walk and carry all the equipment in a backpack. Furthermore, wading doesn't provide a stable surface for various kinds of data gathering and recording.

During a two year-period, we studied the northern leopard frog (*Rana pipiens*) in the Saint-Lawrence river floodplain, Quebec, Canada. In the process we characterized the habitat of tadpoles, from eggs to metamorphosis. Environmental variables included conductivity, dissolved oxygen and underwater light regime, which required carrying heavy and costly equipment in the field. Nutrient and phytoplankton measurements meant that large and heavy volumes of waters had to be carried across the muddy and mosquito's infested floodplain for further analyses to the lab.

To carry this material in the field we used a small inflatable pool boat, which also provided a stable surface to work on (Fig. 1). These are easily found in any major mall. We enhanced the floatability of our small boat and hardened the bottom surface by fixing a small styrofoam surf board at the passenger's place. The total cost of this equipment was about $50. This represents a cheap investment for weekly use, during two tadpoles growing seasons.

However, the following criteria should be considered when selecting a pool boat for wetland studies: 1) The thickness and the sturdiness of the vinyl should provide shock resistance in order to cope with sunken trunks and branches which can easily damage low quality material. 2) The presence of two or more separated air chambers prevents rapid sinking in case of punctures. 3) The inside part of the boat should be deep enough to prevent equipment from falling into the water. With the surfboard, the depth of our pool boat was about 25 cm (8 inches), which was deep enough to keep all the material in. 4) The total inflated size of the boat should allow for easy transportation between the lab and the field; these boats are vulnerable to blowing of the top of a car; so it's better to put it inside a vehicle or in the back of a pick-up. 5) Finally, many models have plastic oarlocks on each upper side. Ropes were attached to these rings and used to pull the charged boat. This offered a purchase which reduced stress on the boat. In addition, we recommend two people moving the boat during field sampling i.e. one pulling and one at the rear to keep an eye on the payload and to push when crossing dense emergent vegetation.

The small pool boat used to carry equipment and samples during our wetland ecological study proved to be extremely useful. It makes the work easier and other laboratories have followed our example. It's a cheap and efficient way to access tadpole's habitats. Why don't you just try it ?

Acknowledgements. We thank J.-F. Lapierre, D. Deshaies, M. Lefevbre, P. Thibeault and G. Martineau for their great help in the field. Thanks to J-F Desroches, F.W. Schueler and ___________ for comments, which improved the manuscript.

REF:

Houlahan, J.E., C. S. Findlay, B. R. Schmidt, A. H. Meyers, and S. L. Kuzmin. 2000. Quantitative evidence for global amphibian population decline. Nature 404: 752-755.

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

Fig. 1. The small pool boat used to carry our equipment in the flooded silver maple forest.

ANNEXES

ANNEXE A

PLANCHE D'IDENTIFICATION DES STADES DE DÉVELOPPEMENT DES LARVES ET TÊTARDS D'ANOURES

Tirée de : Gosner, K.L. 1960. Herpetologica. 16 : 183-190.

Table de développement pour les anoures, développée par Gosner (1960). Stades 1 à 25 : Embryon et larve. Source : McDiarmid et Altig 1999

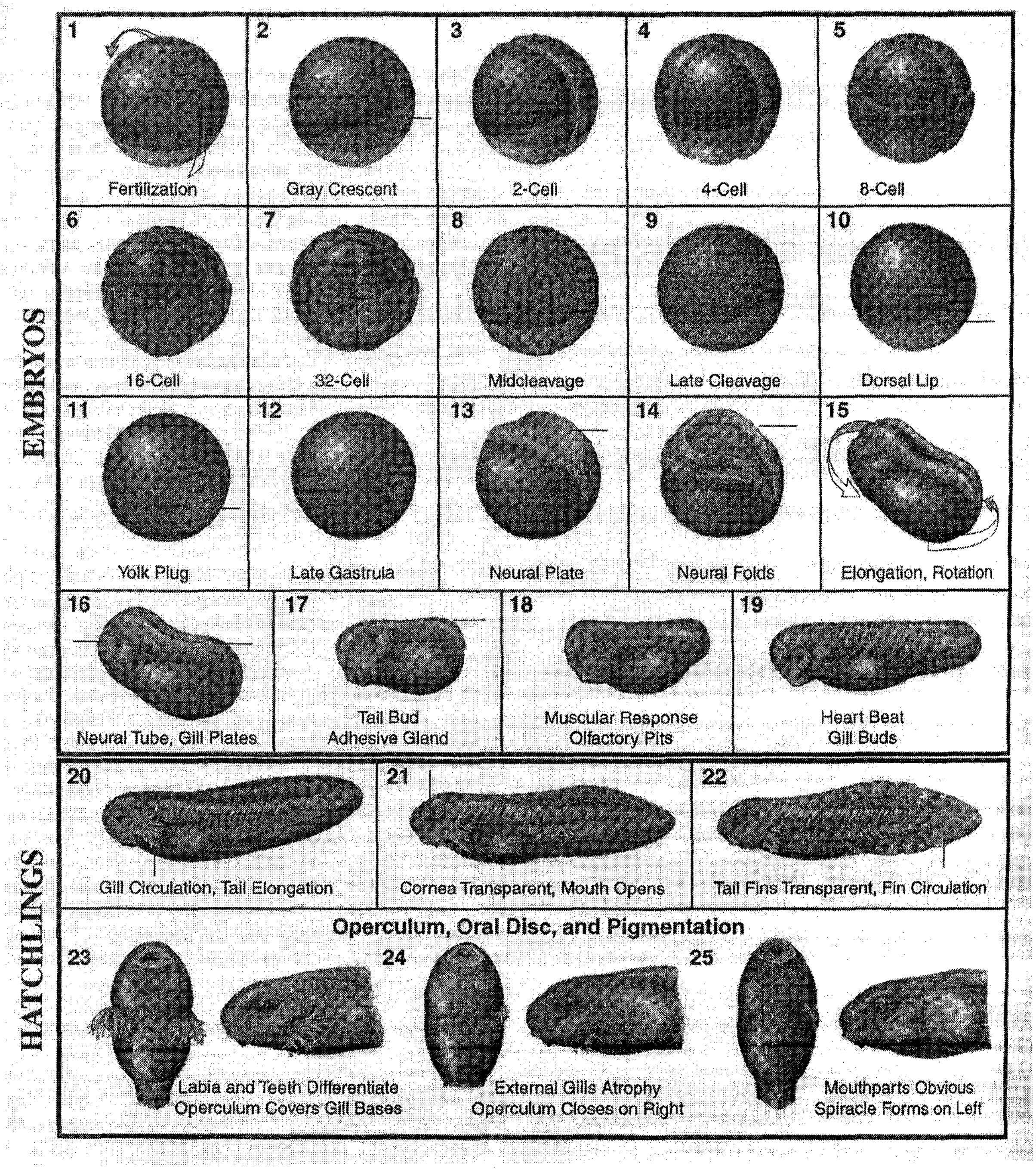
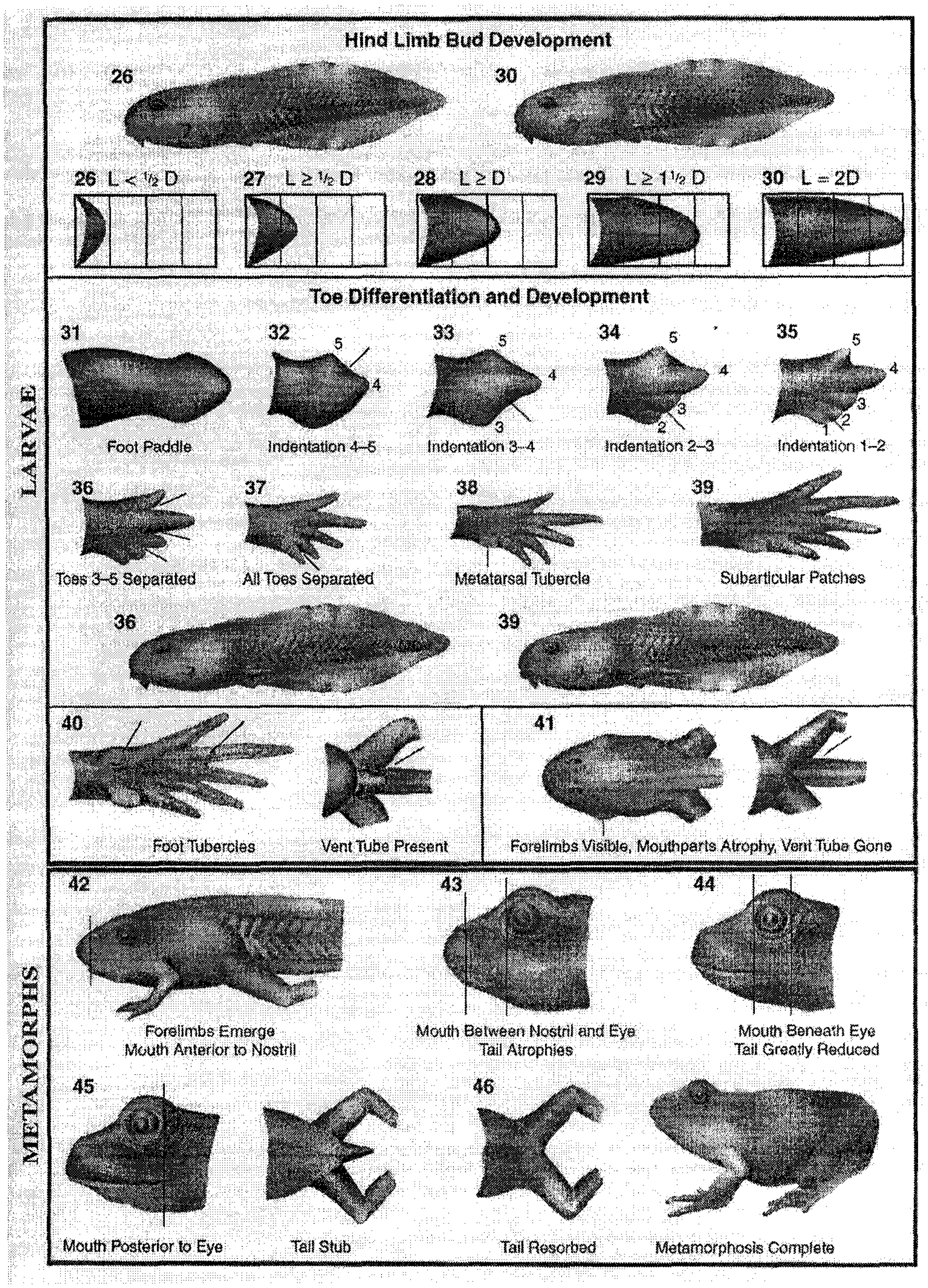


Table de développement pour les anoures, développée par Gosner (1960). Stades 26 à 46 : Têtards et Métamorphes. Source : McDiarmid et Altig 1999.



ANNEXE B

RECOMMANDATIONS AUX AUTEURS

Society for Conservation Biology

Instructions to Authors -- updated: December 2005

Acceptable Manuscripts

The submission rate for Conservation Biology continues to grow, and a large number of manuscripts is handled each year. Given the immutable laws of mathematics, this means less time can be spent on each paper, and many papers must be rejected. But many of the manuscripts received (approximately 25-35%), although of high quality, clearly are not appropriate for this journal and simply waste the time of the authors and the editorial staff. Consequently, greater self-culling is desirable. Here are some types of papers that we typically do not publish unless they have some additional features that argue for their inclusion:

autecological studies of single species or groups of species;
purely descriptive studies that do not address any particular conservation question;
status and trend reports of species, regardless of how dire their conditions might be;
geographic patterns of genetic diversity in a species, with no larger conservation or genetics question addressed;
reports on species distributions and decline;
studies that do not have a conservation question at the core.

Furthermore, species endangerment by itself does not qualify a paper as appropriate for this journal; there should be more substantive content than a descriptive analysis of an endangered species. Before submitting a paper, authors should ask themselves whether the work transcends the particular species or system. Does it address larger conservation questions? If so, are these questions the core of the paper or simply contained in a final couple of paragraphs that discuss "conservation implications?"

Authors should ask themselves if there is much of a chance that a person in a different field or different part of the world might be interested in reading their paper. If only a few specialists are likely to read it then it probably belongs in a more specialized or regional journal.

Manuscript Categories

Conservation Biology accepts submittals for the following categories of manuscripts. Number of words includes all text from the Abstract through the Literature Cited; it does not include tables or figure legends. Manuscripts that significantly exceed the word count will be returned without review.

Contributed Papers (approximately 3000 to 7000 words). Typical papers reporting research projects.

Research Notes (no more than 3000 words). Shorter, sometimes more preliminary research papers.

Review Articles (no more than 7500 words). Comprehensive reviews of a particular topic.

Essays (no more than 7000 words). Analytical papers that are more speculative and less documented than research papers.

Conservation in Practice (no more than 5000 words). Papers that relate experiences in the application of conservation principles to problem solving.

Conservation and Policy (no more than 2500 words). Papers that address the intersections and relationships of conservation science with appropriate policy issues.

Comments (no more than 2000 words). Refers to material previously published in this journal, and usually written as a critique or follow up.

Diversity (no more than 2000 words). Short opinion pieces.

Letters to the Editor should be short (<1000 words). Communications regarding topics of immediate interest to readers, including observations on controversial subjects, on previously published papers, or on other items of note.

Book Reviews are by invitation. All book review manuscripts and communications about book reviews should be sent directly to the book review editor.

Note that the submittal rate to Conservation Biology is increasing, competition for journal space is intense, and criteria for acceptance are strict. Many manuscripts submitted are not appropriate for Conservation Biology. See comments above under "Acceptable Manuscripts."

Manuscript Submission and Specifications (please follow exactly)

All manuscripts must be submitted electronically as Microsoft Word for Windows attachments to an email message. They must be in proper format for a Microsoft Windows or DOS operating system computer. Files written on an Apple MacIntosh system must first be converted to Windows format. All figures must be readable by Word and embedded at the end of the manuscript or submitted together in a separate attachment in one TIFF or EPS file. Tables must be included within the Word document, not as separate attachments. They should follow the Literature Cited and precede Figure Legends. A cover letter (stating the intended manuscript category) should be attached as a separate Word file. Do not use zipped files unless absolutely unavoidable. There should be, at most, three attachments to your submittal email: a cover letter, a manuscript, and possibly a separate file containing figures. Entitle each with the last name of the first author, followed by the content (e.g., SmithLetter.doc or SmithManuscript.doc). The body of the email may be left control or indicate that a manuscript is being submitted.

The subject line should be "Manuscript Submittal." Submit electronic manuscripts to manuscripts@conbio.org (Do NOT send copies to our other email addresses.). Only manuscripts should be submitted to that address; all other correspondence with the editorial office should go to mflagg@conbio.org. If technological limitations prevent submitting a manuscript electronically, you may prepare the manuscript as above and mail an appropriate medium (3.5" diskette, Zip disk, or CD) to Gary Meffe, Editor, Conservation Biology, Wildlife Ecology and Conservation, Newins-Ziegler, Box 110430, University of Florida, Gainesville, FL 32611-0430, U.S.A.

The Conservation Biology "Style Guide for Authors" contains detailed information on how to write and format a paper for Conservation Biology. The document is available on this Web site. Please adhere to its specifications and the following important points.

Use ragged right margins as opposed to justified right margins, and clearly lineate paragraphs by indenting the first line. Avoid large control spaces (such as between sections), and do not use footnotes. Metric measurements must be used unless English measurements are clearly more appropriate, in which case metric equivalents must be given in parentheses. Statistical terms and other measures are to conform to Scientific Style and Format: the CBE Manual for Authors, Editors, and Publishers, sixth edition. Spelling should follow the Webster's Third International Dictionary, and other style points should follow The Chicago Manual of Style, fifteenth edition. We discourage the use of acronyms in the text unless they are absolutely necessary. Pages, including tables, should be numbered at the bottom. Do not number figure pages. . Manuscripts must be in English; U.S. rather than British spelling should be used. We encourage authors whose first language is not English to have a native English speaker edit the paper before submission. A cover page should include the title of the paper, a running head (a shortened version of the title of less than 40 characters), a list of 5 to 8 key words or phrases, word count (all text from Abstract through Literature Cited but not tables or figure legends), authors' addresses at the time the research was conducted and present address(es) including street address (and zip plus four if address is U.S.), and the name and complete mailing address and email of the person to whom correspondence and proofs should be sent.

Abstracts

Each Contributed Paper, Research Note, Review, Essay, or Conservation in Practice article should have an abstract of no more than 300 words (200 for a Research Note). The abstract should state concisely the goals, methods, principal results, and major conclusions of the paper (i.e., should be a "miniversion" of the paper). Incomplete and uninformative descriptions (e.g., "a new method of analysis is given") are not acceptable. Acronyms are not permitted in the abstract. Do not provide a Spanish translation of the abstract.

Citations, Tables, and Figures

Literature citations in the text should be as follows: (Buckley & Buckley 1980b; Pacey 1983). Reference strings should be in chronological order (oldest first). Do not cite unpublished material in Literature Cited (a submitted manuscript is not published and should be cited only in the text as "unpublished data"). The Literature Cited section must be double-spaced. For abbreviations and additional details consult the BIOSIS List of Serials, the CBE Manual, and recent issues of Conservation Biology. Tables must be double-spaced, without vertical rules, and should not duplicate any material in the text or figures. All tables are to have complete but brief headings, be on separate pages, and be numbered consecutively within the text following Literature Cited. Figures and photographs should each be on a separate page. Computer-generated graphics must be of very high quality, with sharp, black line and lettering of a size suitable for reduction. Line drawings should be done in India ink. Lettering should be uniform among the figures. All figures and lettering should be capable of 66-50% reduction without loss of clarity or legibility.Figure legends should be typed double-spaced together on a separate page just before the figures. The combined number of supporting elements (figures and tables) in a manuscript should not exceed a ratio of one element per four pages of text (including Literature Cited).

Additional Submittal Information

Authors are encouraged to provide the names, addresses, and emails of up to four persons qualified to review the manuscript, who have no close working relationships with the authors. The identity of reviewers will be kept confidential unless they choose to be revealed. Authors who are not sure whether their manuscript is suitable for Conservation Biology may send an Abstract to the editor via email (gmeffe@conbio.org) for preliminary evaluation. Include the abstract in the body of the message, not as an attachment.

Policy on Duplicate Publication of Research Results

Submission of a manuscript to Conservation Biology implies that it has not been published previously and is not being considered for publication elsewhere. At the time of submission, authors must describe in a cover letter any data, figures, or text in the manuscript that have been used in other papers that are published, in press, submitted, or soon to be submitted elsewhere. If any of the data in the manuscript have been included in other published or unpublished manuscripts, the legend of each table or figure reporting such data should cite those manuscripts. All manuscripts and authorships are expected to conform to the SCB member code of ethics, available on this Web site.

Page and Color-Printing Charges

Voluntary page charges will be assessed for each paper accepted for publication. Rates are $150 per page for those with grant or institutional support for publication costs and $50 per page for those without support who are willing to pay at this reduced rate. Page charges will be waived for those who will sign a statement to the effect that they have neither institutional support for publication costs nor can they afford $50 per page. An author's ability to pay will in no way influence whether his or her paper will be accepted for publication. Do not include page charge information in your correspondence with the editor. There is a nonwaivable US$1400 per page charge for color figures. Submittal of color figures should be accompanied by a statement in the cover letter recognizing the charges for color and acknowledging an ability to pay.

Reviewing Page Proofs

The copyediting of Conservation Biology is done through the publisher, Blackwell Publishing. Typeset proofs, however, are NOT checked word for word; thus, it is the responsibility of the primary author of each paper to review copyedited manuscript and page proofs carefully for accuracy of citations, formulae, etc., and to check for omissions in the text. It is imperative that the author does a prompt, thorough job of reviewing proofs. It is in the author's power to save himself or herself and the journal the embarrassment of having to explain mistakes that could have been avoided.

Advice for Contributors to *The Canadian Field-Naturalist*

Content

The Canadian Field-Naturalist is a medium for the publication of scientific papers by amateur and professional naturalists or field-biologists reporting observations and results of investigations in any field of natural history provided that they are original, significant, and relevant to Canada. All readers and other potential contributors are invited to submit for consideration their manuscripts meeting these criteria. The journal also publishes natural history news and comment items if judged by the Editor to be of interest to readers and subscribers, and book reviews. Please correspond with the Book Review Editor concerning suitability of manuscripts for this section. For further information consult: A Publication Policy for the Ottawa Field-Naturalists' Club, 1983. *The Canadian Field-Naturalist* 97(2): 231-234. Potential contributors who are neither members of *The Ottawa Field-Naturalists' Club* nor subscribers to *The Canadian Field-Naturalist* are encouraged to support the journal by becoming either members or subscribers.

Manuscripts

Please submit, **to the Editor,** in either English or French, **three** complete manuscripts **written in the journal style.** The research reported should be original. It is recommended that authors ask qualified persons to appraise the paper before it is submitted. All authors should have read and approved it. Institutional or contract approval for the publication of the data must have been obtained by the authors. Also authors are expected to have complied with all pertinent legislation regarding the study, disturbance, or collection of animals, plants or minerals. The place where voucher specimens have been deposited, and their catalogue numbers, should be given. Latitude and longitude should be included for all individual localities where collections or observations have been made.

Print the manuscript on standard-size paper, **doublespace throughout,** leave generous margins to allow for copy marking, and **number each page.** For Articles and Notes provide a citation strip, an abstract and a list of key words. Generally, words should not be abbreviated but use SI symbols for units of measure. The names of authors of scientific names may be omitted except in taxonomic manuscripts or other papers involving nomenclatural problems. "Standard" common names (with initial letters capitalized) should be used at least once for all species of higher animals and plants; all should also be identified by scientific name.

The names of journals in the Literature Cited should be written out in full. Unpublished reports and web documents should not be cited here but placed in the text or in a separate Documents Cited section. List the captions for figures numbered in arabic numerals and typed together on a separate page. Present the tables each titled, numbered consecutively in arabic numerals, and placed on a separate page. Mark in the margin of the text the places for the figures and tables.

Check recent issues (particularly Literature Cited) for journal format. Either "British" or "American" spellings are acceptable in English but should be consistent within one manuscript. **The Oxford English Dictionary, Webster's New International Dictionary** and le **Grand Larousse Encyclopédique** are the authorities for spelling.

Illustrations

Photographs should have a glossy finish and show sharp contrasts. Electronic versions should be high resolution. Photographic reproduction of line drawings, **no larger than a standard page,** are preferable to large originals. Prepare line drawings with India ink on good quality paper and letter (don't type) descriptive matter. Write author's name, title of paper, and figure number on the lower left corner or on the back of each illustration.

Reviewing Policy

Manuscripts submitted to *The Canadian Field-Naturalist* are normally sent for evaluation to an Associate Editor (who reviews it or asks another qualified person to do so), and at least one other reviewer, who is a specialist in the field, chosen by the Editor. Authors are encouraged to suggest names of suitable referees. Reviewers are asked to give a general appraisal of the manuscript followed by specific comments and constructive recommendations. Almost all manuscripts accepted for publication have undergone revision—sometimes extensive revision and reappraisal. **The Editor makes the final decision** on whether a manuscript is acceptable for publication, and in so doing aims to maintain the scientific quality, content, overall high standards and consistency of style, of the journal.

Special Charges — Please take note

Authors **must share in the cost of publication** by paying $80 for each page, plus $15 for each illustration (any size up to a full page), and up to $80 per page for tables (depending on size). Authors may also be charged for their changes in proofs. Reproduction of color photos is extremely expensive; price quotations may be obtained from the Business Manager. If grant or institutional funds are not available, club members and subscribers may apply for a waiver of charges for the first five pages.

Limited journal funds are available to help offset publication charges to authors with minimal financial resources. Requests for financial assistance should be made to the Editor when the manuscript is accepted.

Reprints

An order form for the purchase of reprints will accompany the galley proofs sent to the authors.

FRANCIS R. COOK, Editor
RR 3 North Augusta, Ontario K0G 1R0 Canada

Society for the Study of Amphibians and Reptiles

Herpetological Review is published by the Society for the Study of Amphibians and Reptiles (SSAR), the largest international herpetological society. Membership in SSAR is reasonably priced. Members receive the Society's primary technical publication, the Journal of Herpetology , and its news-journal, Herpetological Review. Both are published four times per year (March, June, September, December). Members also receive pre-publication discounts on other SSAR publications, which are advertised in Herpetological Review. To join SSAR, send payment to: Dr. Theodora Pinou, SSAR Treasurer, Western Connecticut State University, Department of Biological and Environmental Sciences, 181 White Steet, Danbury, CT 06810, USA, Voice (203) 837-8791 and FAX: (203) 837-8769; e-mail: PinouT@WCSU.edu . An extensive list of titles available from the Society may be checked at our Publications page.

Information for Contributors to Herpetological Review

Herpetological Review is a peer-reviewed quarterly that publishes, in English, articles and notes of a semi-technical or non-technical nature, as well as book reviews, institutional features, commentaries, regional and international herpetological society news, and letters from readers directed to the field of herpetology. Articles reporting the results of experimental research, descriptions of new taxa, or taxonomic revisions are not published in HR, but should be submitted to the Journal of Herpetology (http://www.ssarherps.org/pages/JHinstr.php).

Standard Manuscripts (including Articles, Techniques, Herpetological Husbandry, Recent Population Changes, Herpetological History, Points of View, Letters to the Editor). Please send these directly to the Editor (Robert Hansen; rwh13@csufresno.edu).

Electronic submission is strongly encouraged to expedite reviews and reduce costs to the society. Manuscript files should be sent as e-mail attachments in RTF, MS Word, or PDF format. If file sizes are large (i.e., greater than 1 MB), please use a standard compression utility (e.g., zip) to reduce file size before sending via email. Questions about any of this should be directed to the Editor.

Style and Formatting

1- Double-space the entire ms., including the lit cites.

2- Do NOT include an abstract, as HR does not publish these.

3- Do pay careful attention to proper lit cite format, as this is typically the biggest problem area and only results in publication delays and aggravated editors.

4- If English is not your primary language and you think your manuscript could benefit from a pre-review process, please check this link to SSAR's Presubmission Manuscript Review service (http://www.ssarherps.org/pages/presub.html).

5- Illustrations should be discussed in the text and numbered sequentially with Arabic please read numbers. References to illustrations should be placed in parentheses at the end of sentences. (Also, "Graphics Materials/Art Files" below for details concerning how to send figures).

Editorial Conventions

1-For legends, use Fig. 1, Table 3 (no bolding, small caps, and Fig. rather than Figure).

2-0800 h and 24 h (note spacing).

3-Temperatures as 24°C (note spacing).

4-Sample size use upper case N (not italicized), and N = 7 (note spacing).

5-Literature citation for HR is Herpetol. Rev.

6-Spacing items: 78 ± 2.6; P = 0.56; 6-20%; mid-April; 0.5 m

7-Alphabetize references within a series, separated by a semi-colon: (Aaronsen 1955; Burger 1923; Parker 1972).

8-Spell out all state/province names (e.g., Arizona, not AZ).

9-Include country in all postal addresses (e.g., Berkeley, California 94720, USA).

10-Avoid use of personal/professional titles in Acknowledgments (e.g., We thank John W. Jones rather than Dr. John W. Jones).
11-A period should be followed by a single space.
12-Examples of proper literature citation formats:

For an article in a journal or serial publication.: Smith, J. W. 1988. Distributional notes on amphibians of eastern Texas. Texas J. Sci. 42:12-14.

For a book or monograph: Conant, R., and J. T. Collins. 1991. A Field Guide to Reptiles and Amphibians of Eastern and Central North America. 3rd ed. Houghton Mifflin Co., Boston, Massachusetts. 450 pp.

For an article or chapter within a book: Auffenberg, W. L., and W. W. Milstead. 1965. Reptiles in the Quaternary of North America. In H. E. Wright, Jr., and D. G. Frey (eds.), The Quaternary of the United States, pp. 557-568. Princeton Univ. Press, Princeton, New Jersey.

For further guideline, authors should consult Scientific Style and Format: The CBE Manual for Authors, Editors, and P ublishers (6th edition, 1994, Council of Biology Editors, Inc., 11 South LaSalle Street, Suite 1400, Chicago, Illinois 60603, USA).

Animal Care and Permits: The Society feels strongly that all animals used in research be treated humanely and ethically. The Society has published a guide (jointly issued by ASIH and HL) for appropriate treatment of amphibians and reptiles used in field research, and all contributors to HR are expected to comply with these guideline. In addition, HR requires a statement indicating that authors have complied with all applicable institutional Animal Care guideline, and that all required state and federal permits have been obtained. This statement should be included with your manuscript submission cover letter and should also be referenced in the ms. "Acknowledgments."

Graphics Materials/Art Files: Illustrative material (e.g., tables, photographs, diagrams, etc.) should be sent with initial manuscript submissions as low-resolution digital files (as PDFs or JPEGs) or imbedded as figures within the ms. text file. Please do not send original figures at time of ms. submission. If your manuscript is accepted for publication, you have two options: 1) you can send originals via US Mail or courier (with appropriate precautions taken to ensure delivery without damage) and we will take care of the scanning, or 2) you can submit these materials electronically. Download "Guideline for Submission of Digital Art Files" in (pdf) or (html) for specific digital file requirements.

Book Reviews

Review copies of books should be sent directly to the Book Review Editor (Aaron Bauer, Department of Biology, Villanova University, 800 Lancaster Ave, Villanova, PA 19085; email: aaron.bauer@villanova.edu).

News Items, Meeting Listings

Please send meeting announcements, news items, and related materials to the Editor (Robert Hansen; rwh13@csufresno.edu) well in advance of the desired publication date (e.g., by 1 January for the March issue).

Reprints

Paper and/or PDF reprints of notes or articles published in HR should be ordered using the form sent to authors as the issue goes to press. Reprint order forms are not sent to authors of Geographic Distribution notes; these authors should indicate (at time of submission) whether reprints are desired and if so, the appropriate order form will be sent prior to publication. Although it is possible for authors to purchase reprints after the initial order deadline, the prices from Allen Press rise substantially.

HR Review Staff : Editor: Robert W. Hansen: rwh13@csufresno.edu
Snail-mail to: Editor, Herpetological Review 16333, Deer Path Lane Clovis, CA
93611-9735 USA OR: fax 559-327-6590. tel 559-323-7170

Managing Editor:
Thomas F. Tyning: ttyning@berkshire.rr.com

Associate Editors:
Brian W. Bowen: bbowen@hawaii.edu
Robert Espinoza: robert.e.espinoza@csun.edu
Michael Grace: mgrace@fit.edu
Brad Hollingsworth: bhollingsworth@sdnhm.org
Steve A. Johnson: johnsons@wec.ufl.edu
Daryl R. Karns: karns@hanover.edu
Gunther Koehler: gkoehler@senckenberg.de
Deanna H. Olson: Dede.Olson@orst.edu
Christopher A. Phillips: chrisp@inhs.uiuc.edu
Robert N. Reed: reed@suu.edu
Paul A. Stone: pstone@ucok.edu

Section Editors:
Book Reviews: Aaron M. Bauer: aaron.bauer@villanova.edu
Current Research: Eli Greenbaum elig@ku.edu and Maria Castaneda mrcasta@gwu.edu
Geographic Distribution: Jerry D. Johnson: jjohnson@utep.edu
Geographic Distribution: Hidetoshi Ota: ota@sci.u-ryukyu.ac.jp
Geographic Distribution: Alan M. Richmond: alanr@bio.umass.edu
Geographic Distribution: Gustavo J. Scrocchi: soniak@unt.edu.ar
Herp. Husbandry: Ruston Hartdegen ruston17@yahoo.com
Natural History Notes: James H. Harding hardingj@msu.edu, turtles
Natural History Notes: Marc P. Hayes mhayesrana@aol.com, lizards, crocodilians, Sphenodon
Natural History Notes: Charles Painter cpainter@state.nm.us, amphibians
Natural History Notes: Andrew Holycross holycross@asu.edu, snakes
Zoo View: James B. Murphy jbmurphy2@juno.com