ORIGINAL ARTICLE

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Intraspecific competition in tadpoles of *Rana arvalis*: does it matter in nature? A field experiment

Abstract Tadpole growth and development are easily affected by intraspecific competition in tank experiments, provided treatment density is sufficiently high. Is this a phenomenon also observed in nature? A pond was divided into four tadpole-proof sections. Each year for 8 years, all spawn laid by moorfrogs (*Rana arvalis*) in this pond was relocated to create relative spawn and tadpole densities of 1:4:1:4. No direct effect of the density manipulation on survival, tadpole size, and development and metamorph timing and size could be demonstrated. However, I also measured actual tadpole density during the time of development. Apart from the experimental density manipulation, this measure included effects of between-year variation in amount of spawn, natural tadpole mortality, and pond drying (which concentrated the tadpoles by decreasing the area of the pond sections). Actual density had limited but significant effects on tadpole size and development. I suggest that density regulation, acting on the tadpole stage, may be present in the population but was of less short-term importance than abiotic factors and, possibly, adult density regulation. Consequences of the findings for conservation are discussed.

Key words Complex life cycle · Density dependency · Frogs · Growth rate · Metamorphosis · Population regulation

Introduction

Population regulation and frogs

Most people probably agree that density-dependent factors ultimately affect natural animal populations (Lack 1954; Klomp 1962; Gurevitch et al. 1992). However, for most populations, it is not obvious what processes are involved and what is the relative importance of density-dependent and density-independent factors for year-to-year population variation (van Buskirk and Smith 1991). For some groups, e.g., most frogs, an extra dimension is added by the presence of a complex life cycle. In any single population, density-dependent factors may well affect only one of the life stages (Wilbur 1980).

For frogs, studies of density regulation have almost exclusively concerned the aquatic stage. Laboratory studies usually do show effects of density on tadpole survival and performance [Wilbur 1976 (*Ambystoma maculatum, A. laterale, Rana sylvatica*), 1977a (*Rana sylvatica*), 1977b (*Bufo americanus*); Harris 1987 (*Notophtalmus viridescens*); Cummins 1989 (*Rana temporaria*); Murray 1990 (*Rana sylvatica*); Loman 1999 (*Rana temporaria*)]. However, laboratory studies only set a limit to what is possible; they give no conclusive information on what is actually going on in nature.

Field experiments

Most studies detecting the effect of a factor on animal performance take the form of controlled laboratory experiments. There is a problem with this approach. The problem is not with the efficiency of the method to answer the question; it is with the question posed. Often we are not really interested to know if variation in a factor (say, temperature) affects animal performance (say, survival). We are interested in knowing if temperature variation experienced in natural situations affects performance. In a laboratory experiment we can indeed work with "natural" (in contrast to extreme) magnitudes for the effect factors. However, it is impossible to ensure that all the factors we cannot control for (but which may interact with temperature) are at a natural level; to assure this, the simplest approach is to analyze correlations of natural data. However, this is a weak method if one is actually interested in causation.

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The solution to this difficulty is the field experiment. In a field experiment, manipulation is done on animals living in their natural setting in all respects except the factor under manipulation. This approach solves the problem of the effect of background factors. However, it does not automatically solve the problem as to what constitutes natural levels of the test factor; this must be solved by subjective judgment. If this condition is deemed reasonable, it also means that one can put reasonable confidence in the conclusions.

The use of field experiments in tadpole studies

A short review of the use of different "habitats" used for the study of competition in amphibian larvae is given by Scott (1990). He points out the advantages and disadvantages with the different approaches. Basically, they consist of these classes: small laboratory tanks, large outdoor tanks, field enclosures (fenced-off portions of natural ponds), and full ponds. In my opinion, only the last category can be considered field experiments in the strictest sense. The use of artificial ponds (= large tanks) has been criticized by Jaeger and Walls (1989). In defence, Hairstone (1989) and Wilbur (1989) have pointed out the advantages of tanks, provided one is aware of their limitations.

Scott (1990) mentioned ponds only as part of correlative studies. Some other studies based on experiments in natural ponds, used in full (van Buskirk and Smith 1991; Beebee et al. 1996), were also published after the Scott (1990) paper. The present study takes a similar approach.

The approach of this study

Thus, this experiment was designed to avoid some of the shortcomings of more restricted housings when analyzing density effects on tadpole performance. I performed the study under a realistic background level for nonmanipulated factors such as predation, food, and temperature by analyzing tadpoles with unrestricted access to a full natural pond. I used levels of the experimental factor, density, that in each year were close to the natural density of the study population. To accomplish this, the study was done on tadpoles in their native pond. All manipulated densities are of the same magnitude as the natural ones (at most, 60% above).

Part of this study previously was the topic of a preliminary report (Loman 1997), which included data from 5 years. The finished study, which is reported here, comprises data from 8 years. The previous report was based only on measurements of tadpoles whereas the present report also includes information on metamorph measurements. The analysis is now also extended to include more density mea-

sures, and the conclusions have been modified in the light of the added data and analyses.

Methods

Study animal and study pond

The study is concerned with the moor frog *Rana arvalis* Nilss., which belongs to the group of brown frogs in the genus *Rana*. It breeds explosively in spring, in the study area usually during one of the first 2 weeks in April. Adult frogs breed for the first time at 2 or 3 years of age (Loman 1978). Spawn is deposited collectively. Groups of spawn may contain up to 100 clumps, occasionally even more (Loman 1996); much smaller groups are also found. There may be several breeding sites in a single pond. Breeding at one site usually lasts for less than 3 days. Hatching takes place after 10–20 days, depending on the temperature (personal observation).

The study pond is situated in the Revinge area, a military training field in southernmost Sweden (55°40′ N, 13°30′ E). The study pond is about 10 by 100 m in area. Part of it is overgrown by *Salix*. The rest of the pond has a luxuriant growth of submerged and emergent plants such as marsh cinquefoil (*Comarum palustre*) and bullrush (*Typha latifolia*). The pond is temporary and dries up completely in most summers. Following warm and dry springs, this drying takes place before the time for tadpole metamorphosis.

In addition to moor frogs, common frogs (*Rana temporaria*) were also breeding in small numbers in the pond (Table 1). There were ample populations of predators in the pond. Among the more common were the newts *Triturus vulgaris* and *Triturus cristatus*, larvae and imagines of Dytiscidae (e.g., *Dytiscus marginalis*), and leeches (*Haemopis sanguisuga*).

The pond is typical for breeding ponds of moor frogs in southern Sweden. Most moor frogs breed in shallow ponds that more or less frequently dry up in the summer. It is common for them to breed together with common frogs.

Design of the experiment

Before the start of the experiment, the pond was physically subdivided into four similar sections, from east to west, labeled A, B, C, and D. The division was done with a plastic sheet that reached from the bottom of the pond to just above the water level in spring. It was not possible for tadpoles to pass through the sheet. Total conformity between the sections could not be guaranteed, but the proce-

Table 1. Number of spawn clumps found in the study pond

	1990	1991	1992	1993	1994	1995	1996	1997
Rana arvalis	184	108	48	202	130	68	168	251
R. temporaria	21	0	24	15	13	6	2	8

dure and analysis adopted were designed to compensate for this limitation. However, in particular the pairs A and D and B and C, respectively, were quite similar with respect to size and vegetation. A predator inventory also gave similar results for all sections.

After breeding, all spawn deposited by frogs in the pond was redistributed into four new groups of spawn, one in each section. The relative amount of spawn in the four sections after this was $1:4:1:4$. Thus, an initial total number of 100 clumps in the pond, for example, would be relocated as $10:40:10:40$. After that, the eggs were left to hatch naturally and the tadpoles grew in their respective sections, at the densities determined by the number of breeding frogs, experimental manipulation, and, finally, natural mortality in the sections. The procedure was repeated for 8 years (1990–1997). In 1990, 1992, 1994, and 1996 sections A and C were used as high-density sections and B and D were low-density sections. In other years, the section categories were reversed. In 1996, sections A and B were dry at breeding time and all spawn was redistributed to sections C and D in the proportion $4:1$.

There are two reasons to perform the experiment for several years (Polis et al. 1998), apart from the benefit of increasing the sample size. First, it is possible, not to say likely, that the effect of increasing tadpole density depends on the original density. A study spanning several years is likely to give examples of naturally low-density and naturally high-density years. One hopes that the latter approaches the limit set by natural regulation (K in the conventional models based on the logistic equation), whichever the stage it operates on. The second reason is the necessity to compensate for the inevitable difference in the properties of the four sections. Without between-year variation, 2 years would be sufficient to accomplish this. However, it might be that in a dry year section A is superior to D while in a wet year the reverse is true. In other words, there may be an interaction between section and year effects on tadpole performance. Performing the experiment for several years reduces the variation in the response that results from such effects. Complete correction for all factors that might interfere with the planned design of the experiment is not possible, but such are the terms of a field experiment (Morin 1998).

Spawn of *R. temporaris*, which amounted to less than 10% of that of moor frogs, was treated and redistributed in the same way as that of *R. arvalis*. These species have a similar ecology and probably compete (Lardner 1995). Eggs of the two species hatch at the same time and metamorphose at about the same time. It was therefore thought that leaving the spawn of *R. temporaris* at its original breeding places would, in a variable way (because there were different amounts of spawn between years and sections), affect the competitive situation for *R. arvalis* and thus introduce an unwanted noise in the analysis. However, if any competitive effects were found with the design chosen, the main part of it would be intraspecific because *R. arvalis* tadpoles always dominated heavily (see Table 1).

When deciding on the proportion of spawn allocated to the sections, the following considerations were made. It was not useful to arrange a large difference between high- and low-density sections. Increasing density too much above the natural level would not give evidence of the kind of response in which I am interested. Possible effects on the performance of tadpoles after artificially increasing the density tenfold would not prove that any regulating mechanisms found operated at the present natural level. Also, too drastic reduction of density in the low-density sections would make it difficult to obtain samples of tadpoles for measurement. On the other hand, too small a difference between high- and low-density sections would make it impossible, with reasonable sample sizes, to detect any real differences between the two types of treatment. The proportion $1:4:1:4$ was judged a suitable compromise.

Deposited spawn

The natural density variation is the background against which the following results should be judged. Total amount of spawn varied between 72 and 259 clumps per year, most of it from *R. arvalis* (see Table 1).

Practical procedure of manipulation and sampling

The pond was checked every 3–5 days during the breeding period in spring to find all spawn deposited. When a breeding site was detected, it was left intact for 2–3 days to make sure that all egg laying had finished. After that, all spawn (from both *Rana* species) was collected with a sieve. The spawn was weighed and put back in the pond in the determined proportions at four new sites, one in each section. As new breeding sites were found, spawn was likewise collected and redistributed at the same four sites, which were situated similarly to those where the spawn was originally found.

In the middle of May (interyear variation, 17–24 May) and at the beginning of June (3–5 June), samples of tadpoles were taken. In each section and period, 5–7 samples were taken with a scraper net. The total bottom area covered with each sample was $1\,m^2$. If very few tadpoles were found, additional nonsystematic attempts were made to increase the sample for size measurements. As these were not used for density estimates, sample sizes given in Table 2 do not always correspond to the catch data shown later in Fig. 2.

The tadpoles were preserved in 70% alcohol and later measured. Measures (to 0.1 mm) were body length (snout–vent) and hindleg length. Hindleg length was divided by body length to yield an index of development, a tadpole with long legs relative to body being close to metamorphosis. It was also possible, using a preparation microscope, to separate *R. temporaria* from *R. arvalis* tadpoles.

At the time of metamorphosis, samples of metamorphs were caught on the shore by hand, measured (body and tail length, to 0.1 mm), and released. Metamorphs were used for size measurements so long as traces of the tail were visible. Apart from the size measurement, these captures gave information on time for metamorphosis of each individual (Loman 1999), which was considered to have taken place on the day the tail was between 2% and 33% of body length. If the tail was between 33% and 100% of body length, the day

Table 2. Section treatments and sample sizes for data on body size and relative leg length (rel. leg.)

Year	Section	Density treatment	May tadpoles		June tadpoles		Metamorphs	
			Body	Rel. leg	Body	Rel. leg	Size	Day
1990	A	H	56	56	47	47		
	B	L	32	32	47	47	6	6
	C	H	78	78	51	51	4	4
	D	L	8	8	9	9		
1991	A	L	5	5	30	30	22	8
	B	H	2	0	51	51	58	35
	C	L	2	0	14	11	33	15
	D	H	1	0	2	2	40	14
1992	A	H	3	3	22	22		
	B	L	1	1	11	11		
	C	H	2	2	0	0		
	D	L	0	0	0	0		
1993	A	L	0	0				
	B	H	4	4				
	C	L	0	0				
	D	H	0	0				
1994	A	H	47	47	20	20		
	B	L	22	22	18	18	42	17
	C	H	17	17	18	18	60	36
	D	L	14	14	15	15	48	24
1995	A	L	2	2	12	12		
	B	H	2	2	22	22	48	33
	C	L	0	0	13	13	29	22
	D	H	0	0	9	9	54	39
1996	A	H						
	B	L						
	C	H	55	55	36	36	121	90
	D	L	12	12	11	11	89	67
1997	A	L	37	37	38	38		
	B	H	70	70	118	118		
	C	L	26	26	64	54		
	D	H	65	65	52	52		

Blank, section was dry, thus no catch; 0, none were caught and measured, despite water in the section

after this was considered to be the day of metamorphosis. If the tail was longer than the body length, metamorphosis was considered to have taken place 2 days later. Actually, no tadpoles with a tail more than 110% of body length were found on land and measured. Samples were taken every 3–5 days during the period of metamorphosis. The capture distribution was used to compute the median date for metamorphosis for each section and year.

The sample sizes for the different measurements are given in Table 2. Note that metamorphs with a tail less than 2% of body length were not used for estimating day of metamorphosis but, provided traces of tail were still visible, were used for estimating size. Thus, the sample size for size measures was larger than that used for metamorph day measures.

Effects of spring weather and pond area on experiment design

Following dry and warm springs, the pond dried up before time of metamorphosis, or even before the time for the planned second tadpole sample (Fig. 1). During the latter part of tadpole development, differences between years and sections in pond drying rate affected the actual density as much as did variations in spawn deposited.

Measures of density and analysis

Because the drying of the pond by necessity affected the relative density in the four sections and also did so differently in different years, the analysis was carried out at two levels, using three measures altogether. First, I analyzed the direct effect of the treatment (high versus low density) on tadpole (May and June) and metamorph performance with two-way ANOVAs. The factors used are year and density (low versus high). Sections with same density treatment were pooled, because in some years only one low- or high-density section remained water filled at time of sampling (see Fig. 1).

Second, I analyzed the effect of actual density on tadpole and metamorph performance, using ANCOVAs to account for year and density. For May tadpole measures, I used May density. For June tadpoles and metamorphs, I used the average of May and June densities, which was deemed a better measure than June density alone; as tadpole growth had taken place under a variety of densities, the average of these two measures was my best estimate. Actual density was, in turn, measured using two alternatives, section density and local density.

Section density was, for each section, period, and year, defined as the average number of tadpoles per scraper net
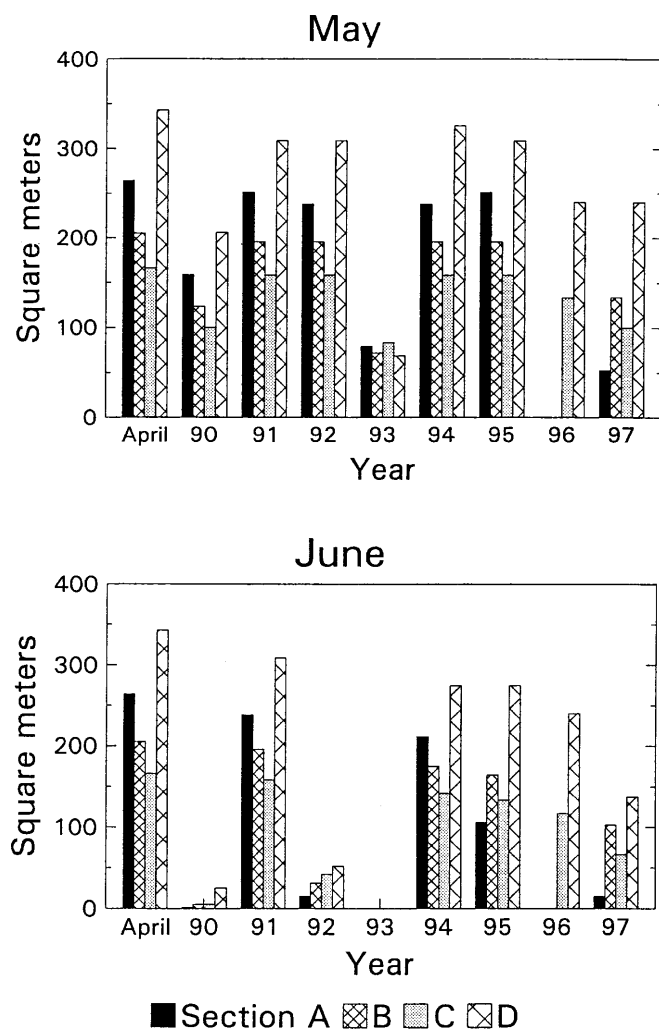
May

June

■ Section A ⊠B □C ⊠D

Fig. 1. Original section areas and areas at the time of tadpole sampling. At the time of spawning in April, the pond area was similar in all years. This area is indicated in the *leftmost cluster* of both parts of the figure. In 1993, the pond was completely dry before the time when the second sample was scheduled. In 1996, sections A and B were already dry at breeding time in April

sample ($1 m^2$ each). As an alternative to section density, I used local density because the sections were not internally homogeneous; sometimes there were considerably more tadpoles captured in some samples than in others (among the 5–7 samples taken in each section–period–year). Presumably, these samples came from parts of the sections that were favored by tadpoles. Therefore, I weighted the sample densities by the number of tadpoles per sample, which gave an estimate of the average density each tadpole captured actually had experienced. If all samples had the same density, section and local density was the same. If some samples had higher densities, the average tadpole could be expected to have been living at a density more close to the higher density measured than to the average sample density. In this case, local density is higher than section density.

The formula for section density can be expressed as

$$\sum_{i=1}^{n} S_i / n$$

and that for local density as

$$\sum_{i=1}^{n} s_i^2 \bigg/ \sum_{i=1}^{n} s_i$$

where s is a sample size and n is the number of samples.

Tadpole performance measures as used in ANOVAs were tested for homogeneity of variance using the Levene's test; this did not demonstrate large and significant violations of the assumption.

Results

In all following analyses, year is entered as a factor. Because year variation is large, it is important to account for this in the search for density effects. The year effect itself is nearly always significant. As this is not considered of interest for the scope of this study, it is not commented on further. The data are, however, given in the tables.

Persistence of the experimental density manipulation

In May, densities in sections designed (by spawn transfer) to have a high density were indeed higher (Fig. 2A). The effect of the treatment was significant [two-way ANOVA (year and density treatment): $df = 1:21$, $F = 9.775$, $P = 0.005$]. In June, section density was not significantly affected by the density treatment [two-way ANOVA (year and density treatment): $df = 1:18$, $F = 1.497$, $P = 0.237$] (Fig. 2B). The fact that the density differences persisted at least until May makes it meaningful to search for density effects on performance of tadpoles (see Performance in high- and low-density sections).

Survival in high- and low-density sections

Despite the fact that density differences between high- and low-density sections persisted in May, an effect of spawn density on tadpole survival from hatching to May could very well be present; this would have reduced the original number of tadpoles by different proportions between high and low spawn sections. The null hypothesis is that the difference in tadpole number remained a factor of four, similar to the difference in egg number. Number of tadpoles was estimated as density (from scraper net samples) times remaining section area. However, an ANOVA testing for May values in which those in the low-density sections were multiplied four times showed no significant effect ($df = 1:21$, $F = 0.728$, $P = 0.403$). Thus, no significant effect on tadpole survival rate from spawn density could be found. Also, for June, no spawn density effect on survival could be demonstrated; testing after multiplying the low spawn density sections June tadpole number by 4 also was not significant ($df = 1:18$, $F = 2.273$, $P = 0.149$).
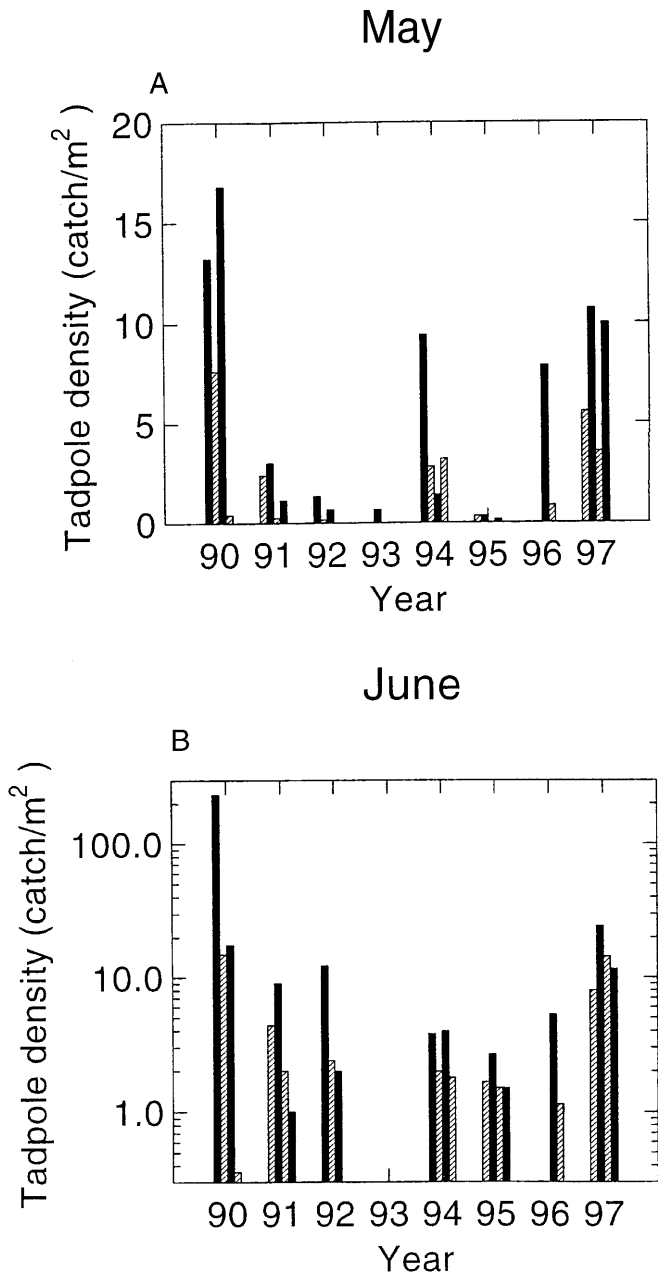
May

A

June

B

Fig. 2. Tadpole section density in May (**A**) and early June (**B**). The *bars* in each cluster represent the four sections. *filled bars*, high-density sections; *hatched bars*, low-density sections. *Missing bars*, no tadpoles were captured; except for the two missing bars in 1996 and all in June 1993, these sections were dry at the time of sampling

Performance in high- and low-density sections

Tadpole body length

In both May and June, there was a significant effect of the interaction between year and density treatment (Table 3; Fig. 3 A,B). Actually, in 1990 and 1994 tadpoles were larger in the low-density section but in 1996 (June) and 1997 the reverse was (surprisingly) true. In the other years, samples were small (May 1992 and 1995; see Table 1), differences were slight, or there were no data at all (1993) as a result of

pond drying. Because the interaction is significant, I did not interpret the main effects.

Tadpole relative leg length

In the May sample, there was no evidence for difference in development rate between the density treatments and there was no significant interaction between year and density (Table 3; Fig. 3C). Also, in June there was no effect of density on development, but the significant interaction effect (Table 3; Fig. 3D) showed that at least in some years density affected development.

Metamorphs

For metamorph data (size and timing of metamorphosis), no effect of density treatment was found; neither the factor density nor its interaction with year was significant (Table 3; Figs. 4A,B).

Performance related to actual (section and local) density

Local density affected body length in May (Table 4). Both section and local density affected tadpole body length and relative leg length in June (Table 4; Fig. 5A,B); tadpoles growing at high densities were smaller and had relatively shorter legs than those growing at low densities. There was no significant effect on metamorph size (Table 4; Fig. 5C), but a marginally significant effect of local density on time for metamorphosis occurred (Table 4; Fig. 5D); tadpoles growing at high densities metamorphosed later than those growing at lower densities.

Discussion

Density effects in this population

I think the results of this study can be summarized as follows. There are indeed density effects on tadpole performance in the field. However, they are comparatively minor relative to the variation among years and also in any one year, among ponds (personal observation). However, in any single year and especially in ponds that are prone to drying during early summer, initial tadpole (egg) density explains only a small part of the variation in tadpole density during the latter part of tadpole development.

Only the second approach gave evidence for competition. Why not the first approach, where direct effects of the treatment were studied? Part of the explanation may be that the first method lost power because of the natural pond drying that randomly perturbed the designed densities. However, these variations usually had little effect on the May densities (but much more in June). On the other hand, in defense of the method, note that tadpoles captured in June had spent most of their growth at densities determined by the experimental design and nonweather-dependent factors. Only during the last 1–2 weeks (between the May and

**Table 3.** Effects of density treatments (high- or low-density section) on tadpole performance

		DENS			YEAR			DENS*YEAR		
		df	F	P	df	F	P	df	F	P
Tad. body	May	1:570	0.000	0.959	6:570	265.650	0.000	6:570	22.340	0.000
	June	1:716	8.614	0.003	6:716	135.256	0.000	6:716	13.222	0.000
Tad. rel. leg	May	1:542	1.663	0.198	5:542	300.844	0.000	*5:537*	*0.974*	*0.433*
	June	1:703	4.761	0.029	6:703	9.552	0.000	6:703	19.530	0.000
Metam. body		1:648	0.046	0.830	4:648	52.294	0.000	*4:644*	*0.495*	*0.740*
Metam. date		1:404	1.421	0.234	4:404	131.154	0.000	*4:400*	*0.611*	*0.655*

The effect was tested with a two-way ANOVA that also accounts for year effects; if an interaction was nonsignificant, it is printed in italics
The test was then rerun without the interaction and those results (for DENS and YEAR) are listed
DENS, density; YEAR, year; *df*, degrees of freedom; Tad. body, tadpole body size; Tad. rel. leg, tadpole relative leg length; Metam., metamorph

Fig. 3. Average measures of tadpoles captured in high-density (*filled bars*) and low-density (*hatched bars*) sections in May and June. **A** Body size in May. **B** Body size in June. **C** Relative leg length in May. **D** Relative leg length in June
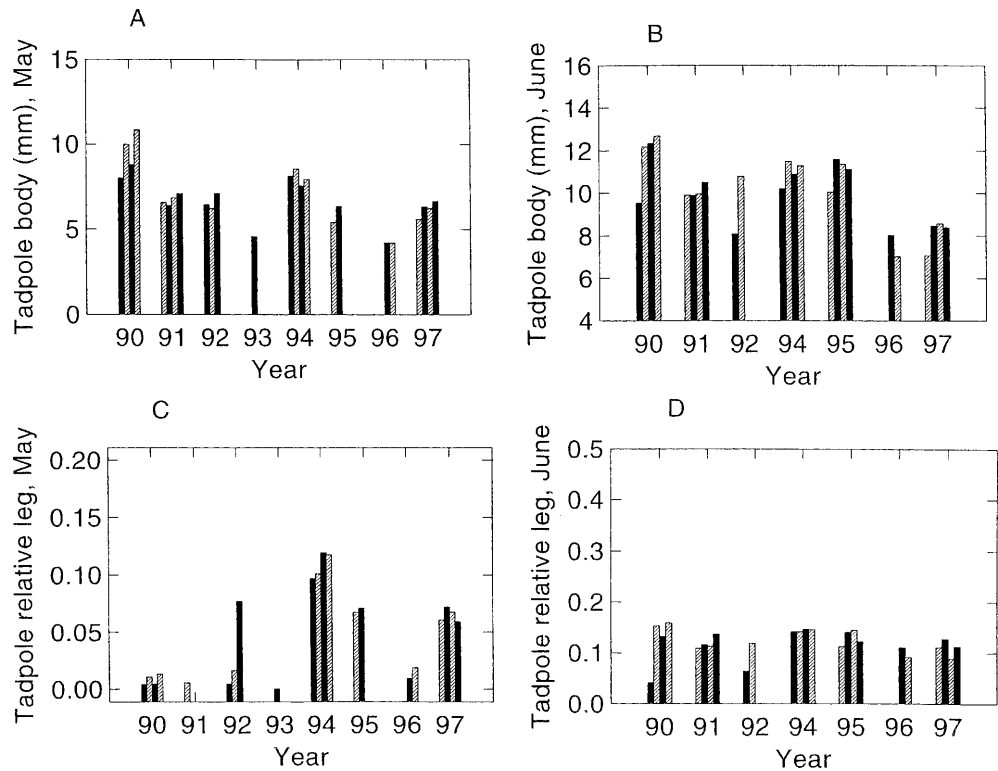
June sample times) in some years did drought have a variable influence on density and survival between sections.

The net conclusion is this. The second analysis (based on actual tadpole density) showed that there is a potential for density regulation, even within the ranges of densities encountered in nature. The lack of a direct effect of the experiment shows that this is only an average effect, likely to be important in the long run only. In any single year, any effects from crowding are more likely to be caused by dry weather than by high density of the breeding frog population. For population regulation to operate on the tadpole stage, there must be a correlation between adult population size and crowding effects in the pond. For ponds in which the area is often affected by drying, such as the present study pond, this correlation may be weak. In populations breeding in permanent ponds, population regulation acting on tadpoles may be more important.

Threshold effects

Disregarding year effects, one can use Fig. 5 to search for threshold effects. Could it be that density regulation influences tadpole performance only in years with very high densities? Actually, only the very high density experienced by some tadpoles in 1990 gave an obvious example of such effects; these seemed to have an arrested development rate (see Fig. 5B). On the other hand, they were actually larger than tadpoles living at lower densities in other years (Fig. 5A). However, this is not a fair comparison; 1990 was a very early year with metamorphosis almost 10 days earlier than in any other year (Fig. 5D), which makes Fig. 5A unsuitable for between year comparisons. The conclusion from Fig. 5B is for the same reason conservative, however. Another suggestion of a threshold effect comes from the metamorph size data (Fig. 5C). In the high-density year 1990, meta-
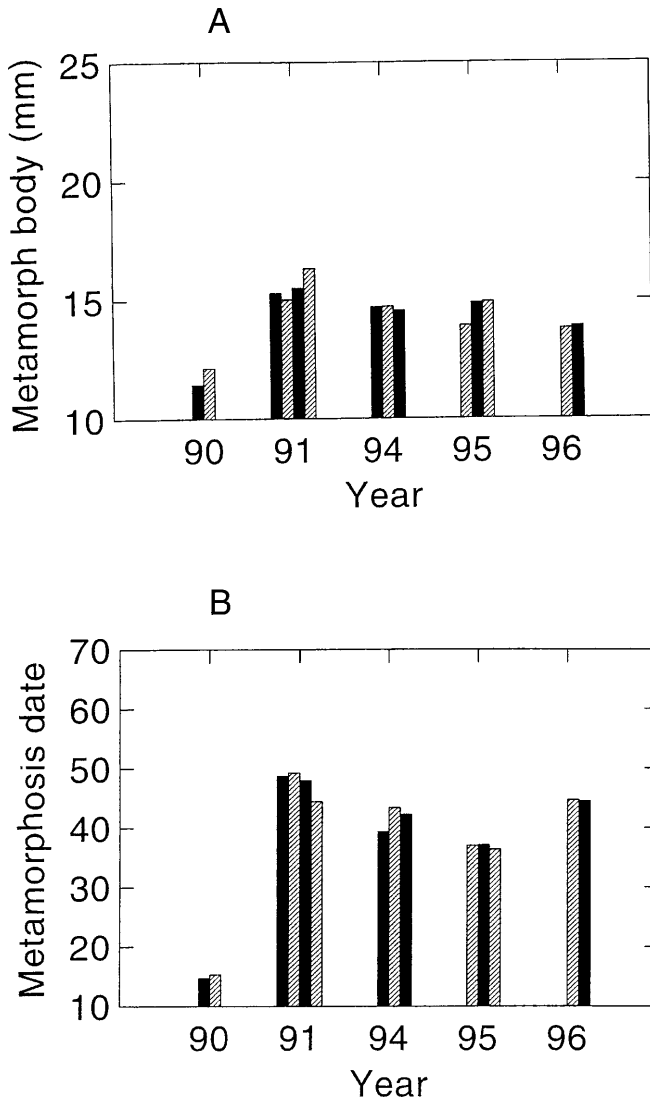
A

B

Fig. 4. Average data for metamorphs captured at high-density (*filled bars*) and low-density (*hatched bars*) sections. **A** Metamorph body length. **B** Time for metamorphosis (days after June 1). In missing years, no tadpoles metamorphosed

morphs were quite small. However, the lack of a within-year effect from density puts this conclusion in question; an alternative explanation could be that the quick development, in turn resulting from the exceptionally warm spring of 1990, was the cause of the small metamorphs.

Problems with the method

The strength of the field experiment method is obvious; any effects demonstrated have, almost by definition, relevance for what actually affects the studied population. The weakness is of course the great variation in contribution of factors that are not fully controlled for. In the present case, one factor should be mentioned. Growth and development rate varied greatly between years. The most serious aspect of weather variation was the variable tendency for pond and section drying, which influenced density; this was controlled for by the experimental approach in which several sections were analyzed in any one year. However, this could not guard against the possibility of an interaction between seasonal weather and overall density. Also, that would level out if a sufficient number of years were analyzed.

Other studies

As stated in the Introduction, laboratory studies consistently have shown density effects on tadpole survival and performance. The results from studies that more closely attempt to mimic natural conditions are, however, not conclusive. These studies utilize field enclosures or small natural bodies of water and attempt to stock them with tadpoles in natural densities.

A lack of, or minor, density effects on tadpole survival and performance have been found for *Rana clamitans* (Calef 1973), *Rana sylvatica* and *Rana pipiens* (DeBenedicti 1974), *Hyla eximia* (Sredl and Collins 1992), *Pseudacris triseriata* and *P. crucifer* (Skelly (1995a), *Bufo calamita* (Beebee et al. 1996), and the salamander *Notophtalmus viridescens* (Harris 1987) (a density effect was, however,

Table 4. Effect of log density (section and local) on tadpole performance

		YEAR			DENS		
		df	F	P	df	F	P
Tadpole body (May)	Section density	7:15	19.534	0.000	1:15	2.842	0.113
	Local density	7:15	22.911	0.000	1:15	5.147	0.038
Tadpole body (June)	Section density	6:16	11.625	0.000	1:16	6.691	0.020
	Local density	6:16	16.304	0.000	1:16	15.535	0.001
Rel. leg (May)	Section density	7:12	8.821	0.001	1:12	0.235	0.636
	Local density	7:12	9.281	0.001	1:12	0.774	0.396
Rel. leg (June)	Section density	6:16	2.113	0.109	1:16	10.545	0.005
	Local density	6:16	16.304	0.000	1:16	15.535	0.001
Metamorph length	Section density	4:8	13.396	0.001	1:8	4.118	0.077
	Local density	4:8	11.983	0.002	1:8	1.826	0.214
Metamorphosis date	Section density	4:8	189.38	0.000	1:8	2.108	0.185
	Local density	4:8	281.46	0.000	1:8	5.327	0.050

The effect was tested with an ANCOVA that also corrects for year effects; all interactions are nonsignificant and were removed before final analysis

Fig. 5. Relation between tadpole and metamorph performance and local density. Tadpole performance was measured in June; tadpole local density is average of May and June measurements. There is one measurement for each section (if available) and year. The *symbols* represent the last digit in the respective year. *Regression lines*, computed yearwise, are included.
A Tadpole body length.
B Tadpole relative leg length.
C Metamorph size. **D** Time for metamorphosis (days after June 1)
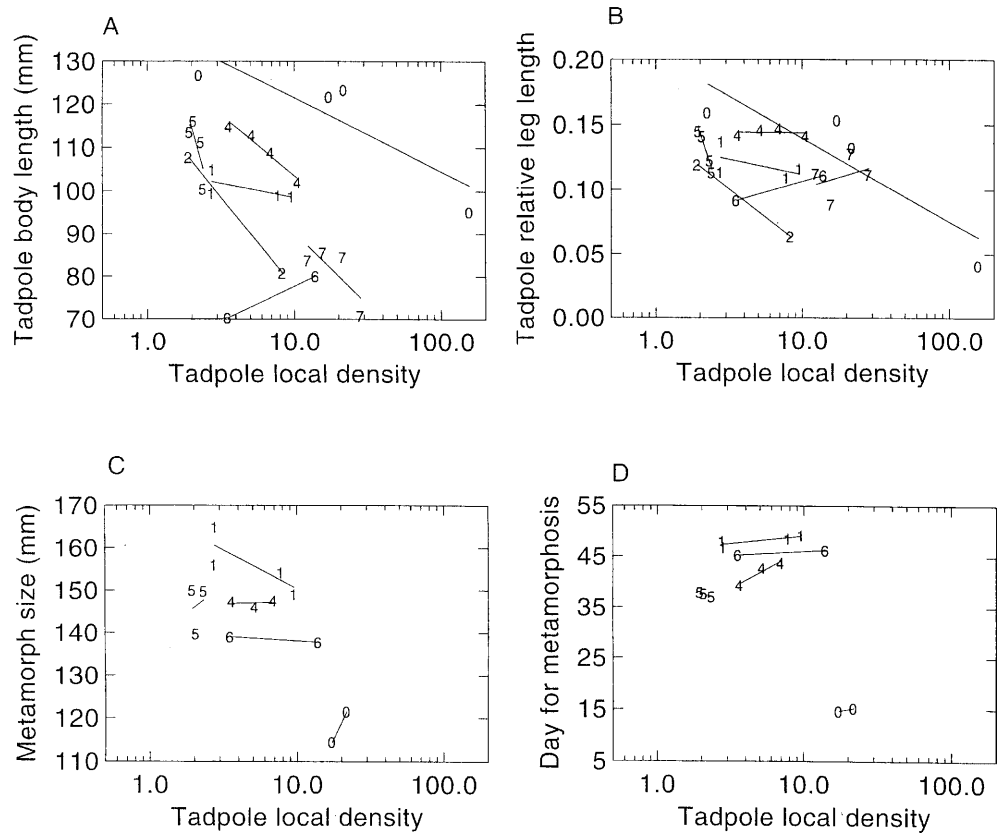
found in laboratory setups). In contrast, Skelly (1995b) found evidence of density regulation in *Rana clamitans*, and similar results have come from descriptive studies in natural ponds [Smith 1983, *Pseudacris triseriata*; Berven 1990, 1995 (one of three sites), *Rana sylvatica*]. Berven (1995) furthermore showed that larval mortality was key to adult population size. Density effects have also been found in salamander larvae (van Buskirk and Smith 1991, *Ambystoma laterale*; Scott 1990, *Ambystoma opacum*). However, this is not immediately comparable to the situation for frog tadpoles, salamander tadpoles being potentially cannibalistic. Further support for the existence of density regulation comes from studies in large outdoor tanks where interspecific competition between tadpoles has been demonstrated (Griffiths 1991; Kupferberg 1997; Parris and Semlitsch 1998). The only (to my knowledge) field experiment of amphibians at the terrestrial stage (Pechmann 1995; studying the salamanders *Ambystoma opacum* and *A. talpoideum*) failed to find an effect on density on survival.

The fact that the conclusions from other studies of this problem differ is not at all surprising but reflects the complex anuran life cycle. In a landscape favorable for adult frogs, populations should increase until pond conditions become limiting for the increasing number of tadpoles. On the other hand, if there is less suitable habitat for adult frogs, these populations may never increase to levels where there is competition between tadpoles. So, even for one species, one is likely to find populations with as well as without tadpole density regulation. Experiments with tad-

poles stocked to natural levels in natural ponds (with or without subpond enclosures) should come to these conclusions. This situation also means, unfortunately, that there is very little generality to expect from these studies; they represent case studies of the conditions in a local population.

Tadpole performance and population density

This study, as well as other similar studies, considers two types of response variables, the first of which is tadpole survival. The importance of this factor for population dynamics is obvious. Second, there are various measures of tadpole and metamorph performance such as tadpole growth rate, development rate (time for metamorphosis), and metamorph size, which are explicitly or implicitly assumed to indirectly affect population dynamics. Indeed, correlative studies on both anurans and urodeles (Smith 1987; Semlitsch et al. 1988) have shown that large metamorphs also are large at sexual maturity, which presumably gives a reproductive advantage. Also, Berven and Gill (1983) found that metamorph size in *Rana sylvatica* correlated positively with juvenile survival. This finding does not prove that tadpoles that grow slowly because of competition, or other adverse conditions during early life, will be smaller as adults or survive less well for this reason. Although this is quite likely, strictly speaking it is also possible that a large size at metamorphosis and as an adult are correlated traits, with no cause and effect involved.

To prove a causation ("silver spoon" effect, sensu Grafen 1988), experimental manipulations must be performed and followed up until the time of the studied response. A short-term study of that sort (John-Alder and Morin 1990) showed indeed that experimentally crowded toads (*Bufo woodhouseii*) grew slower and jumped less persistently. A direct link from tadpole density to adult performance has also been shown by Scott (1994) and by Morey and Reznick (2001). The latter showed that froglets that were small (on a given date) because of small size at metamorphosis usually remained behind in growth until 1 year of age, when the study was terminated. Thus, indeed, it is likely that crowding can affect adult size, which in turn may affect clutch size, for example (Berven 1988; Joly 1991).

In temporary ponds, development rate also has another important implication. If crowding decreases development rate, this means that crowding may have an indirect effect on survival. This is the case if the pond dries during the period of tadpole metamorphosis.

Conservation

The answer to the question, "which is the regulated stage for this population?" has important consequences for conservation. If a population is regulated at the aquatic stage, construction of new ponds should increase population size, but if the population is regulated at the terrestrial phase, one must improve the habitats favored by adult frogs (Wilbur 1980; Beebee et al. 1996; Loman 1997). Presently, the emphasis in frog conservation has been on pond construction. The relatively small density effects in this study and in some others (Calef 1973; Beebee et al. 1996; Skelly 1995a) suggest that this is not always the best option.

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