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PERFORMANCE OF TADPOLES FROM THE HYBRIDOGENETIC *RANA ESCULENTA* COMPLEX: INTERACTIONS WITH POND DRYING AND INTERSPECIFIC COMPETITION

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Abstract.—The performance of three genotypes (LL, LR, RR) of tadpoles resulting from the hybrid mating system of *Rana lessonae* (phenotype L, genotype LL) and *Rana esculenta* (phenotype E, genotype LR) was determined in artificial ponds. The effects of interspecific competition and pond drying on growth, development, and survival of tadpoles were used to measure the performance of genotypes and the relative fitness of offspring. Among the three genotypes, tadpoles from the homogametic mating RR had the lowest survival, growth, and development under all environmental conditions. Body size of the LL and LR genotype tadpoles at metamorphosis was reduced by competition and pond drying. Days to metamorphosis were also higher for the LL and LR genotype tadpoles in competition ponds. The proportion of individuals metamorphosing of each genotype was differentially lowered by competition and pond drying. The LL genotype produced more metamorphs than the LR genotype in the constant water level ponds, but the LR genotype produced more in drying ponds. In competition ponds, the LR genotype produced more metamorphs than the LL genotype, but the LL genotype produced more metamorphs in ponds without competition. The RR genotype produced no metamorphs in any of the experimental environments. Increased performance of LR offspring from the heterogametic mating, in harsh conditions, and reduced performance of RR offspring from the homogametic mating, even under favorable conditions, relative to the parental genotype (LL) suggests that the population dynamics of this hybridogenetic system is strongly dependent on mate choice in mixed populations and the subsequent pond environment females select for oviposition and larval development.

Key words.—Amphibian, development, growth, hybridogenetic, metamorphosis, phenotypic variation, *Rana esculenta* complex, survival, tadpole.

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The *Rana lessonae-Rana esculenta* waterfrog complex (L-E system) is a unique hybrid system (Graf and Polls-Pelaz, 1989). *Rana esculenta* is an interspecific hybrid between *R. lessonae* and *R. ridibunda* with a reproductive mode known as hybridogenesis (Schultz, 1969; Berger, 1968, 1977). During gametogenesis, the hybrid excludes one of its parental genomes, and thus, produces only gametes containing the other parental genome. Because this exclusion is premeiotic, recombination through crossing over is normally prevented and the remaining genome is transmitted clonally (Graf and Polls-Pelaz, 1989). In the diploid L-E system in central Europe (including our study sites in Switzerland) the *R. ridibunda* genome is transmitted and the *R. lessonae* genome is lost. *Rana ridibunda* is no longer sympatric in this system and *R. esculenta* populations can persist only through sexual parasitism, i.e., they must mate with *R. lessonae* to regain the lost genome in each generation. Thus, within the L-E system mixed matings between *R. lessonae* and *R. esculenta* readily occur in most populations, and produce fertile offspring. Frogs of the L-E system are also unique among clonal vertebrate hybrids (e.g., fish and lizards; Lynch, 1984; Vrijenhoek, 1989), because of their bisexual mating system and apparent preference for hybrid mating (Blankenhorn, 1977; pers. obs.).

The relative viability and fertility fitness of offspring produced by hybrid matings is crucial to understanding the evolution and maintenance of hybridogenetic systems (Lynch, 1984). Despite a wealth of information on genetic mechanisms of clonal inheritance and on geographic variation in the expression of hybridogenesis, there are few experimental data on the relative fitness of offspring among environmental conditions. In populations with both *R. lessonae* and *R. esculenta,* there is the potential for four mating combinations that result in four distinct offspring genotypes (Fig. 1). For example, the two heterogametic matings (L ♀ × E ♂ and E ♀ × L ♂) produce highly heterozygous *R. esculenta* offspring. The

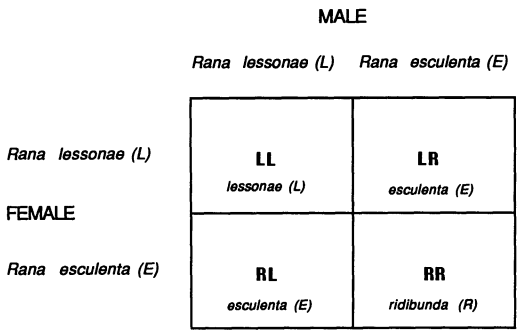


FIG. 1. Mating design, adult phenotypes (L, E), and possible offspring genotypes (LL, LR, RL, RR) obtained from hybrid matings of *Rana lessonae* and *R. esculenta* waterfrogs.

homogametic mating L ♀ × L ♂ produces *R. lessonae* offspring and the E ♀ × E ♂ mating leads to two clonally transmitted *R. ridibunda* genomes and highly homozygous *R. ridibunda* offspring (Berger, 1977; Turner, 1980). Hence, the study of the performance of these genetically different offspring under a range of environmental conditions allows us to determine components of fitness related to viability and fertility, and to evaluate the importance of differential fitness in the selective maintenance of hybrids in populations.

The relative performance of offspring genotypes can be used to determine the consequences of the hybrid mating system in terms of sexual selection if offspring performance is correlated with adult traits related to mate choice (reviewed by Bradbury and Andersson, 1987). Offspring performance can also be used to determine the outcome of natural selection if it is related to viability or fertility of adults in terrestrial habitats (reviewed by Endler, 1986; Travis and Mueller, 1989). In amphibians, recent studies have shown that characteristics related to successful mating such as male body size are positively associated with larval growth rates and size at metamorphosis (Woodward, 1986; Woodward et al., 1988; Mitchell, 1990). Several long-term field studies have also demonstrated that increased size at metamorphosis and early timing of metamorphosis favorably influence age and size at first reproduction, survival to first reproduction, and fecundity (Berven and Gill, 1983; Smith, 1987; Semlitsch et al., 1988; Berven, 1990). Thus, the performance of larval amphibians under a variety of environmental conditions, as measured by growth and development to metamorphosis, can be used to determine adult fitness.

Consequently, we wanted to know the following: (1) Do offspring resulting from specific mating combinations, within a single population, differ in their performance when subjected to complex aquatic environments? (2) Does the performance of these offspring vary depending on the environmental condition in which they were reared, that is, does offspring genotype interact with environmental condition? And (3) can offspring performance in seminatural artificial ponds be related to the differential distribution of phenotypes found among natural breeding ponds?

MATERIALS AND METHODS

Experimental Design and Treatments

We measured the effects of pond drying and interspecific competition on the performance of three genotypes of tadpoles (LL, LR, RR) that result from natural matings between *Rana lessonae* and *R. esculenta* (Fig. 1). Populations of tadpoles were subjected to two types of water conditions: a constant water level to simulate a permanent pond, and a gradually declining water level to simulate an ephemeral pond. Tadpoles were also reared with or without interspecific competitors to simulate the extremes in competitive environments that can be found among natural breeding ponds. All three factors were crossed in a 3 × 2 × 2 (genotype × drying × competition) factorial design yielding 12 treatments and were replicated in three randomized complete blocks of 36 artificial ponds.

We used fiberglass tanks (1.04 m wide × 1.47 m long × 0.80 m deep) as artificial ponds. Tanks were randomly positioned in a rectangular array of three spatial blocks in a fenced field at the university. Blocks were used to partition variation in sunlight across the field. Each of the 12 treatments was randomly assigned within each block. Tanks were filled with 1,100 liters of tap water between 31 March and 2 April 1990. All tanks then received 1.0 kg of air-dried reeds collected along the edge of a natural

pond and were then covered with lids made of fiberglass window screen (1 mm mesh) on 3 April. Tanks were inoculated with 1.0 liters of concentrated plankton collected from six different natural ponds on 3, 9, and 18 April. We also added four adult snails (Lymnaeidae) to each tank on 29 April.

The competition treatment consisted of the presence or absence of two species of tadpoles (*Hyla arborea* and *Bufo calamita*) to simulate a complex competitive environment. *Hyla arborea* occur in a broad range of pond types from permanent to ephemeral, are active swimmers, and feed primarily on phytoplankton. It metamorphosed after an average of 57.4 days at 447 mg body mass. *Bufo calamita* occur most commonly in ephemeral ponds, are bottom dwellers, and feed primarily on algae and detritus. It metamorphosed after an average of 46.6 days at 105 mg body mass. Eggs and tadpoles of both species have been found in the same natural ponds as *R. lessonae* and *R. esculenta* (pers. obs.), but the extent of competition among species is unknown. Clutches of eggs of both species were obtained from ponds near Bulach, Kanton Zürich, Switzerland and were hatched in the laboratory. We added 35 *H. arborea* and 100 *B. calamita* tadpoles to half the experimental ponds on 6 May and 8 May, respectively by species. These represent realistic densities (0.123 tadpoles/liter) to determine experimentally larval responses to competitors (Morin, 1983; Wilbur, 1987).

The drying treatment began on 21 June 1990 in half the tanks by natural evaporation and by draining water with a movable standpipe. Rate of drawdown followed a negative logistic drying curve of a natural pond, and water level was adjusted according to the curve every six days. The drawdown stopped just before complete drying to allow the census of all remaining tadpoles after 60 days. The 60-day experimental hydroperiod was within the range of natural pond hydroperiods, especially disturbed sites. Rapidly developing tadpoles were capable of metamorphosing after just 37 days, and all surviving individuals metamorphosed from constant water level ponds with no competition by day 60, thus providing the standard for the experimental hydroperiod. Constant water level in the other tanks was maintained by rainfall and the single addition of 4 cm of tap water on 25 July. Water level in each tank and minimum-maximum water temperature in two drying tanks and in two constant water level tanks was measured each week.

Adult frogs (10 pairs *R. lessonae* and 10 pairs *R. esculenta*) were obtained on 25 May 1990 from a small, isolated pond near the Katzensee in Kanton Zürich, Switzerland. Each frog was identified to species by enzyme (albumin) electrophoresis and then paired into four possible mating combinations of male and female *R. lessonae* and *R. esculenta* (Fig. 1). Five pairs of each combination were placed in artificial ponds (1.0 m wide × 1.0 m long × 0.8 m high with screen sides and a cover) filled with 20 cm of water. Rocks were provided for male calling sites and aquatic macrophytes were provided for female oviposition sites in each pond. Between 6–12 June 1990 we collected four clutches of LL, three clutches of LR, and three clutches of RR offspring. No eggs were obtained from the RL mating combination. Eggs were allowed to hatch in the laboratory and clutches within each mating combination were pooled together. Tadpoles of each type were haphazardly counted into groups of 20 tadpoles and then two groups of 20 tadpoles of the same genotype (total of 40 tadpoles) were randomly added to each experimental tank on 21 June 1990 (day 0), according to the design.

Measurement of Tadpole Resources

To determine the relative impact of interspecific competition from *Hyla* and *Bufo* on *Rana* tadpoles we estimated resource levels in each tank. Because tadpoles, especially *Rana,* are scrapers that feed on algae and periphyton attached to the surface of vegetation and objects in natural ponds we estimated the resource level by measuring the amount of biomass that accumulated on experimental glass slides. On 15 May 1990 six glass microscope slides (26 mm × 76 mm) were attached to the side of each tank (north side) approximately 20 cm below the surface of the water. On 25 June, four days after adding *Rana* tadpoles we randomly selected two slides from each pond to estimate resource level. This was when *Hyla* and *Bufo* competitors were metamor-

TABLE 1. Summary of body size at metamorphosis (mg), days to metamorphosis, number of metamorphs, body size (mg) and developmental stage of tadpoles, and number of tadpoles from three genotypes of tadpoles reared in replicate artificial ponds.

Treatments				Metamorphs			Tadpoles		
Genotype	Water level	Competition	Rep.	Body size	Days	Number	Body size	Stage	Number
LL	Constant	No	I	872.9	50.2	33	1,781.0	40	2
			II	956.2	48.7	34	2,199.4	41	1
			III	994.0	47.4	37	—	—	0
		Yes	I	353.5	59.0	7	750.7	37	26
			II	462.0	55.3	10	835.2	36	25
			III	492.8	54.7	27	1,094.2	40	5
	Drying	No	I	790.1	55.8	20	1,378.7	39	8
			II	622.7	52.6	23	909.5	38	2
			III	447.6	52.1	31	738.6	39	6
		Yes	I	—	—	0	333.7	28	16
			II	273.3	57.2	12	590.2	36	23
			III	238.1	59.2	5	481.1	35	31
LR	Constant	No	I	731.5	54.2	23	1,293.9	39	14
			II	764.8	45.4	39	3,186.8	31	1
			III	955.0	54.3	12	2,554.7	40	8
		Yes	I	330.0	58.0	6	475.1	34	31
			II	605.1	54.7	26	895.7	39	7
			III	327.2	53.5	11	519.6	35	28
	Drying	No	I	483.3	54.7	25	833.3	38	12
			II	703.7	46.2	36	—	—	0
			III	601.2	48.8	39	—	—	0
		Yes	I	404.2	55.5	19	870.9	38	7
			II	299.0	51.3	21	712.6	38	12
			III	261.3	59.6	5	494.8	35	30
RR	Constant	No	I	—	—	0	55.3	25	11
			II	—	—	0	—	—	0
			III	—	—	0	62.3	25	16
		Yes	I	—	—	0	48.5	25	8
			II	—	—	0	—	—	0
			III	—	—	0	54.2	25	12
	Drying	No	I	—	—	0	42.8	25	1
			II	—	—	0	—	—	0
			III	—	—	0	52.1	25	6
		Yes	I	—	—	0	—	—	0
			II	—	—	0	—	—	0
			III	—	—	0	36.3	25	1

phosing and already had exerted their maximum effect on resources, but before the drying treatment could differentially affect the periphyton community. Slides were air-dried for 48 hours, then periphyton from one side (i.e., the side exposed to tadpoles) was scraped onto preweighed boats and oven-dried for 24 hours at 50°C. Periphyton from the two slides from each pond was then weighed to the nearest 0.1 mg and expressed as mean biomass per cm^2.

Response Variables and Statistical Analyses

Once tadpoles in tanks began to metamorphose, metamorphs were collected from all tanks every two to three days and held in the laboratory until tail resorption was complete. The experiment was ended on 20 August 1990 after *Rana* tadpoles were exposed to environmental conditions for 60 days. On day 60, we drained and thoroughly searched all tanks for any remaining tadpoles or metamorphs. Performance of tadpoles was determined by (1) time to metamorphosis (days from the start of the experiment on 21 June to forelimb emergence; stage 42, Gosner, 1960); (2) size at metamorphosis (body mass to the nearest 1.0 mg after tail resorption; stage 46, Gosner, 1960); (3) percent survival (percentage of all individuals surviving from those ini-

tially added to ponds, including tadpoles); and (4) percent metamorphosis (percentage of survivors that metamorphosed). Mean values per tank were the unit analysis because measurements from individuals within tanks were not independent. Because the RR genotype produced no metamorphs, analyses of metamorphic responses include only two genotypes.

Percentage data were angularly transformed by the arcsine of the square root and body size and time to metamorphosis were logarithmically transformed before analysis to reduce skewness (Snedecor and Cochran, 1980). Differences in treatment effects and their interactions were analyzed by fixed effect univariate analysis of variance (ANOVA) and means were compared by Scheffe's multiple range tests.

Results

Developmental stage and body length of tadpoles at the beginning of the experiment were determined on a subsample ($N = 20$) from each genotype preserved from those added to ponds on 21 June 1990. Tadpoles of all genotypes were at average developmental stage 25 (range 23–25) at the start of the experiment, and stage was independent of genotype ($\chi^2 = 8.59$, $df = 4$, $P > 0.05$). Body length of tadpoles, however, was different among the genotypes ($F = 3.98$, $df = 2, 57$, $P = 0.0241$). A Scheffe's Test indicated that LL tadpoles ($\bar{x} = 4.2 \pm 0.09$ mm) were 7.7% larger than LR tadpoles ($\bar{x} = 3.9 \pm 0.09$ mm), but that RR tadpoles ($\bar{x} = 4.1 \pm 0.02$ mm) did not differ from LL or LR tadpoles. It is unlikely that these small body size differences contributed to significant differences among genotypes during the experiment because these body size differences in hatchlings are discordant with most differences at metamorphosis (see below).

Survival

Total survival was used only as a measure of viability during the larval stage. Because all remaining tadpoles were censused before ponds dried completely, we can separate mortality due to viability from that of desiccation due to pond drying. The survival of all individuals (i.e., metamorphs and tadpoles) was significantly different among genotypes and drying regime (Tables 1 and 2). Total survival of the RR genotype (11.4%) was lower than either the LL (80.0%) or the LR genotype (85.8%). The LL and LR genotypes did not differ in total survival, the proportion of metamorphs and tadpoles surviving in these two genotypes was similar (Table 1). Thus, genotype differences were due solely to the inviability of the RR genotype.

Total survival in drying ponds (54.3%) was marginally significantly lower than in constant water level ponds (63.9%; Fig. 2). Only the RR (3.3%) and LL (73.8%) genotypes suffered reduced survival in the drying ponds versus constant water level ponds (19.6% and 86.2%, respectively by genotype; Fig. 2). LR tadpoles did as well in drying ponds (85.8%) as in constant water level ponds (85.8%, Table 1 and Fig. 2). There was no significant effect of competition or of the two-way interactions on total survival (Table 2).

Development and Metamorphosis

The percentage of LL and LR survivors metamorphosing from ponds was significantly affected by interspecific competition, but not by drying regime or genotype (Tables 1 and 2). No metamorphs were produced from any ponds containing the RR genotype, primarily because of low survival during the first two weeks of larval development. Surviving RR tadpoles reached only the average developmental stage 25 (Gosner, 1960) and were small in body size ($\bar{x} = 50.2$ mg), and had no possibility to metamorphose. The LL and LR genotypes produced equal percentages of metamorphs (60.1% and 63.4%, respectively), and the remaining tadpoles were significantly more developed and larger in average size than RR tadpoles (LL— stage 37, LR—stage 37; $F = 75.5$, $df = 2, 18$, $P < 0.0001$; LL— 1,008.4 mg, LR— 1,183.7 mg; $F = 242.6$, $df = 2, 18$, $P < 0.0001$).

Ponds with no competition produced over twice as many metamorphs (85.5%) as ponds with competitors (38.0%). The interaction of genotype with drying regime also significantly affected the percentage of metamorphs (Tables 1 and 2). In constant water level ponds the LL genotype produced more

TABLE 2. Summary of univariate analysis of variance for survival, percentage of survivors metamorphosing, body size at metamorphosis, and days to metamorphosis for three genotypes of *Rana lessonae* and *R. esculenta*. Data were transformed before analysis (see text).

Response variable	Source of variation	*df*	MS	*F*-value	*P*-value
Survival:	Genotype	2	3.4699	69.64	<0.0001
	Competition	1	0.0524	1.05	0.3145
	Drying regime	1	0.1946	3.90	0.0588
	Genotype × comp.	2	0.0017	0.03	0.9672
	Genotype × drying	2	0.0360	0.72	0.4950
	Competition × drying	1	0.0207	0.42	0.5314
	Residual	26	0.0498		
Metamorphs:	Genotype	1	0.0236	0.28	0.6125
	Competition	1	2.3089	26.85	0.0001
	Drying regime	1	0.0325	0.38	0.5532
	Genotype × comp.	1	0.1099	1.28	0.2740
	Genotype × drying	1	0.4329	5.04	0.0384
	Competition × drying	1	0.0397	0.46	0.5132
	Residual	17	0.0859		
Body size:	Genotype	1	0.0003	0.04	0.8452
	Competition	1	0.5736	68.25	<0.0001
	Drying regime	1	0.1488	17.71	0.0007
	Genotype × comp.	1	0.0063	0.75	0.4090
	Genotype × drying	1	0.0105	1.25	0.2803
	Competition × drying	1	0.00001	0.00	0.9929
	Residual	16	0.0084		
Days:	Genotype	1	0.0005	0.70	0.4236
	Competition	1	0.0114	15.63	0.0011
	Drying regime	1	0.0006	0.83	0.3844
	Genotype × comp.	1	0.0002	0.21	0.6606
	Genotype × drying	1	0.0016	2.27	0.1511
	Competition × drying	1	0.00005	0.07	0.7965
	Residual	16	0.00073		

metamorphs (70.9%) than the LR genotype (57.1%), but in drying ponds the LR genotype produced more metamorphs (69.8%) than the LL genotype (49.2%; Fig. 3).

Body Size at Metamorphosis

Body size at metamorphosis was significantly affected by both competition and drying regime (Tables 1 and 2). Metamorphs emerging from noncompetition ponds were twice as large (743.6 mg) as those from competition ponds (367.9 mg; Fig. 4). Metamorphs from constant water level ponds were larger (653.8 mg) than those from drying ponds (465.9 mg; Fig. 4). There was no effect of genotype on the body size of metamorphs, although only two genotypes produced metamorphs. There were also no significant two-way interaction effects on body size (Table 2).

An analysis of covariance for body size at metamorphosis, using total survival as the covariate, also indicated significant effects of competition ($F = 96.5$, $df = 1, 15$, $P < 0.0001$) and drying regime ($F = 27.9$, $df = 1, 15$, $P = 0.0001$), but not for genotype or any of the two-way interactions.

Timing of Metamorphosis

Days to metamorphosis were significantly affected by competition (Tables 1 and 2). Metamorphs in competition ponds took longer to reach metamorphosis (56.2 days) than those from noncompetition ponds (50.9 days; Fig. 5). Neither genotype nor drying regime nor any two-way interactions significantly affected days to metamorphosis (Table 2; Fig. 5).

Physical Condition and Resource Level in Ponds

Water level in drying ponds declined from 67 cm on 21 June to 5 cm on 20 August. Water temperature in all ponds averaged 14.0°C (minimum) and 20.0°C (maximum) on 21 June. Water temperature in drying ponds (13.0°C minimum and 30.2°C maximum) became more variable than water

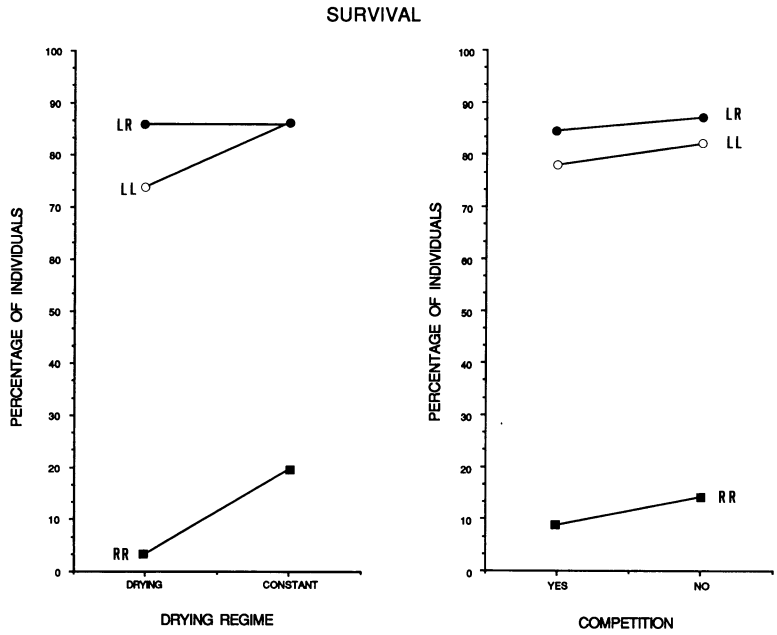


FIG. 2. Percentage of total survivors from three genotypes (LL, LR, RR) of tadpoles reared in two competitive environments and two drying regimes. Values plotted are the means from six ponds.

temperature in constant water level ponds (16.2°C minimum and 23.8°C maximum) by 18 August.

Biomass of resources in the artificial ponds was significantly affected by the competition treatment (Table 3). Biomass in the ponds with *Bufo* and *Hyla* tadpoles (2.7 mg per cm^2) was significantly lower than biomass

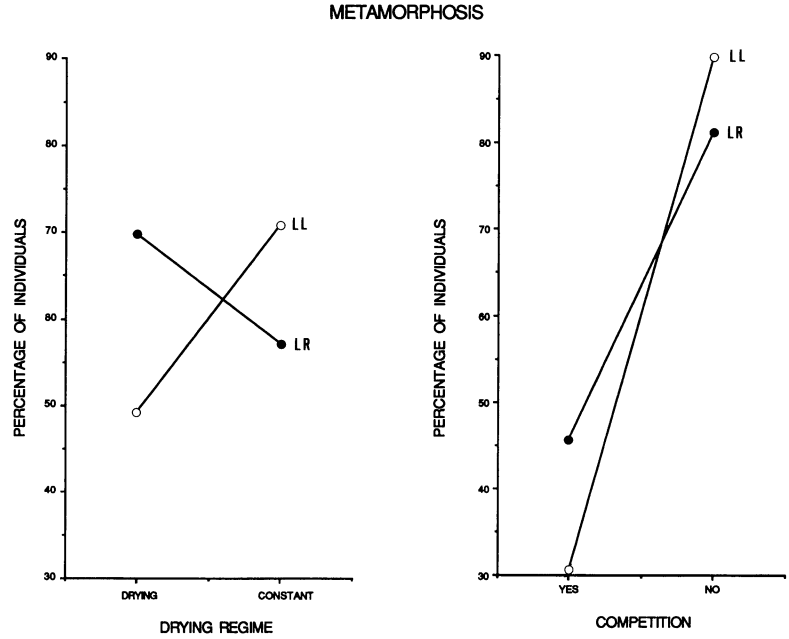
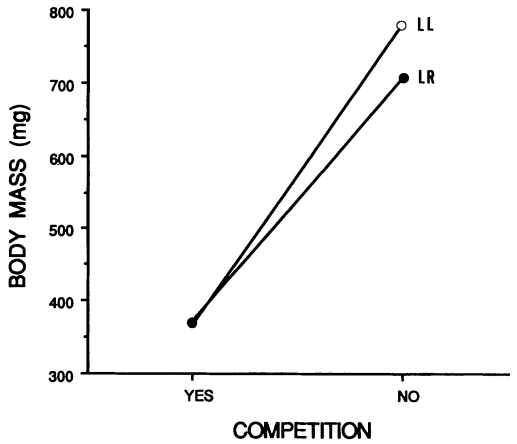


FIG. 3. Percentage of survivors that metamorphosed from two genotypes (LL, LR) of tadpoles reared in two competitive environments and two drying regimes. Values plotted are the means from six ponds.



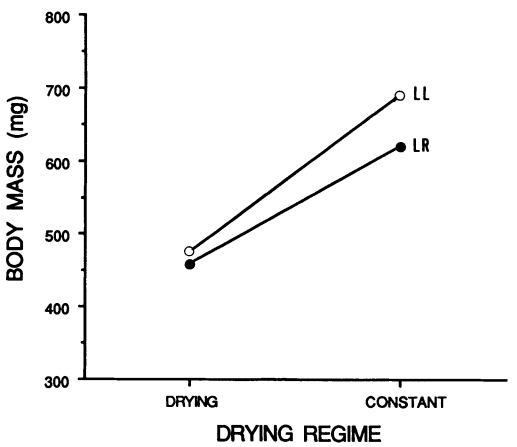
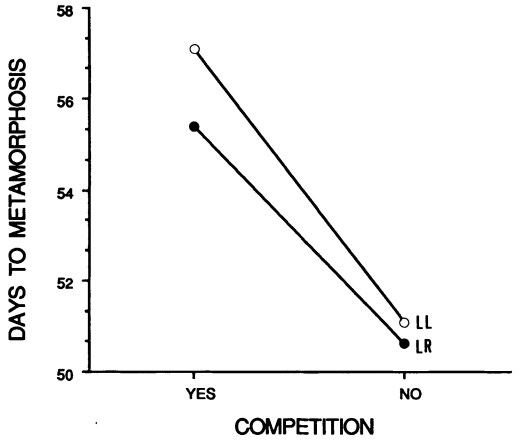


FIG. 4. Body size at metamorphosis (mg) for two genotypes (LL, LR) of tadpoles reared in two competitive environments and two drying regimes. Values plotted are the means from six ponds.



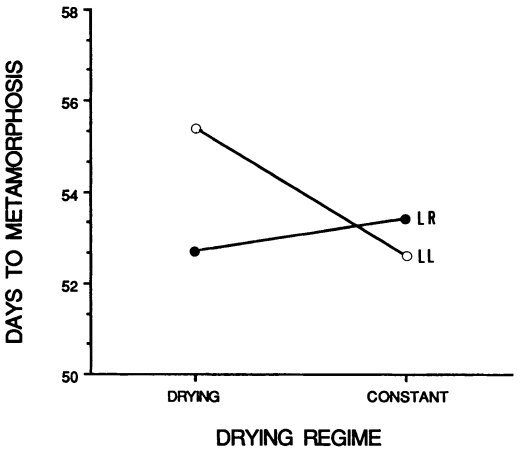


FIG. 5. Time to metamorphosis (days) for two genotypes (LL, LR) of tadpoles reared in two competitive environments and two drying regimes. Values plotted are the means from six ponds.

levels in ponds without tadpole competitors (43.0 mg per cm^2). This indicated that the reduced performance of *Rana* tadpoles in the competition ponds was likely due to reduced food level.

DISCUSSION

Our results show that the performance of tadpoles from this hybridogenetic system was dependent on both the larval genotype (i.e., mating combination) and the environment. Survival of the tadpoles was strongly dependent on the genotype of larvae. Body size and the timing of metamorphosis were dependent on environmental condition, but the number of metamorphs produced in ponds was determined by the interaction between larval genotype and the environment.

Performance of tadpoles from the homogametic mating (RR, *Rana ridibunda* phenotype) was poor under all four environmental conditions of pond drying and competition. There essentially was no development past stage 25 (Gosner, 1960), the stage at which tadpoles were added to the ponds. This was true in all replicate populations, despite using three sibships from this mating cross. Survival of tadpoles was extremely low and no metamorphs were produced during a 60-day period. Very few tadpoles were seen in ponds after the first week, suggesting that most died early in the experiment.

TABLE 3. Summary of univariate analysis of variance for biomass of resources in artificial ponds in relation to treatments.

Source of variation	*df*	MS	*F*-value	*P*-value
Genotype	2	45.00	0.08	0.9249
Competition	1	14,633.73	25.45	<0.0001
Drying regime	1	282.86	0.49	0.4967
Genotype × comp.	2	51.54	0.09	0.9145
Genotype × drying	2	197.96	0.34	0.7119
Competition × drying	1	321.31	0.56	0.4694
Residual	26	574.96		

The poor performance of RR tadpoles in our study was similar to results from other laboratory studies showing abnormal development and extremely low viability (Berger, 1967, 1973; Berger and Uzzell, 1977). It is thought that clonally transmitted genomes from both parents result in high levels of homozygosity and the accumulation of recessive deleterious mutations (Graf and Polls-Pelaz, 1989). Our results, however, extend these findings to another population within the L-E system and to more natural complex aquatic environments suggesting that the low viability of this homogametic mating is general. Travis (1983) and Mitchell (1990) have both previously noted that differences in offspring performance are stronger in the field than in the laboratory because of the interaction of numerous environmental variables affecting growth and development. Thus, it is unlikely that the RR genotype resulting from a homogametic mating of *R. esculenta* hybrids would contribute significantly to the maintenance of hybrid populations by the production of metamorphs and the adult host species (i.e., *R. ridibunda*) for mating.

Performance of tadpoles from the other homogametic (LL, *R. lessonae* phenotype) and the heterogametic (LR, *R. esculenta* phenotype) matings was significantly higher than that of RR tadpoles. Both LL and LR tadpoles attained the largest body size and had the shortest development time in constant water level ponds with no competition. The drying and competition treatments decreased food availability, subsequently lowering growth and development of individuals. Interspecific competitors clearly reduced periphyton levels in the tanks by more than an order of magnitude, whereas the drying condition presumably decreased the volume of water and the area available for feeding.

The number of metamorphs of each genotype, however, was dependent on the environmental condition. The LL genotype produced more metamorphs than the LR genotype in constant water level ponds with no competitors (87% versus 62%), but not when there were competitors (37% versus 36%). The LR genotype produced more metamorphs than the LL genotype in drying ponds with competitors (38% versus 14%), but both genotypes produced similar numbers of metamorphs in ponds without competitors. Thus, for the response that would have a direct numerical effect on fitness (i.e., number of offspring leaving the pond) the two genotypes differ in performance depending on the pond drying regime.

For each of the other fitness responses (i.e., survival of tadpoles, time to and body size at metamorphosis) and for the effect of competition on the percentage of individuals metamorphosing, genotype differences were less marked and in none of the seven cases statistically significant (Figs. 2–5). In each case, however, the effect of harsh environments (i.e., drying and competition) appeared to be more detrimental for *R. lessonae* than for *R. esculenta.* Here, "more detrimental" means that a shift from benign to harsh conditions seems to decrease tadpole survival, rate of metamorphosis, size at metamorphosis, and increase time to metamorphosis more in LL than in LR tadpoles (cf. slopes in Figs. 2–5). A combined pairwise test on these relative decreases and increases, indeed, shows a significant difference between the two genotypes ($P = 0.02$, $T = 0$, $N = 7$, two-tailed, Wilcoxon matched-pair test). Biologically, such a combination of treatments and responses seems to be jus-

tified, because fitness is determined by the combined effects of several ecological conditions on multiple traits. Consequently, even small differences in tadpole survival as well as in growth and development may add up to substantial differences in fitness.

Thus, we suggest that LL tadpoles perform well under favorable conditions (no competition, constant water level), but poorly in harsh environments, perhaps indicating a narrow range of environmental tolerance and local adaptation. Conversely, LR tadpoles appear tolerant to a broader range of environmental conditions, including competition and pond drying. This latter result correlates well with the finding that *R. esculenta* adults, at least, are found most frequently in disturbed habitats such as gravel pits, which are often subject to drying (Blankenhorn, 1977). In addition, *R. esculenta* is known to be more tolerant to hypoxic stress than either *R. lessonae* or *R. ridibunda* (Tunner and Nopp, 1979).

The lack of a significant three-way interaction among genotype, competition, and pond drying in a preliminary analysis ($P = 0.9606$) indicates that the advantage to LR tadpoles is less than expected from its advantage under each environmental condition separately. Thus, the presence of LR tadpoles in a pond might not be as much a cost to LL tadpoles as expected from individual effects. Nevertheless, our results indicate that in mixed populations of *R. lessonae* and *R. esculenta* the genotype advantage would shift between LL and LR tadpoles depending on the pond and how the environment changes temporally and spatially. Recent evidence suggests that female anurans can choose oviposition sites and thereby influence the environment of their offspring (Resetarits and Wilbur, 1989). Females choosing ponds favorable for development and survival of offspring can increase their fitness through offspring viability.

Fitness of a genotype, however, is not solely the result of offspring performance, but is instead the product of offspring viability and adult fertility. Graf (1986) proposed a model for the population dynamics of the L-E system, which combined mating frequency between species with fecundity. He used an analysis of mating frequencies between *R. lessonae* and *R. esculenta* presented by Blankenhorn (1977), which indicated that small male *R. lessonae* mated preferentially with large female *R. lessonae,* but that large male *R. lessonae* preferentially mated with large female *R. esculenta.* Because adult *R. esculenta* females are larger than *R. lessonae* females and larger females produce more eggs, the L ♂ × E ♀ mating combination would produce more offspring (Berger and Uzzell, 1980). Because of these advantages Graf (1986) predicted that *R. esculenta* would have a higher reproductive fitness and thus, be more successful. If we assume that the heterogametic crosses LR and RL resulting in the *R. esculenta* phenotype are equal in larval performance (unpubl. data), then Graf's model can be extended to include both fitness of the larval stage from our results of differential performance and that of adult reproductive fitness to explain the ecological success of *R. esculenta.*

Yet, the maintenance of hybrid populations in this L-E system is contingent on maintaining reproductive adults of both species. Only metamorphs can contribute to juvenile recruitment and the maintenance of species populations. As we have shown this depends on the relative advantages afforded genotypes among environmental conditions. The hybrid advantage of *R. esculenta* should shift spatially with the pond habitat and potentially the adjacent terrestrial environment, and should shift temporally with changing environmental conditions as ponds undergo alteration or succession, both influencing primary productivity (i.e., food level) and pond hydroperiod (i.e., risk of desiccation). Thus, *Rana esculenta* should increase in population size relative to *R. lessonae* in low productivity and disturbed habitats, but decline in more stable, high productivity sites as *R. lessonae* increases in population size. Optimal equilibria between the two species, of course, would depend on how the density and frequency of each affected mate choice. How these factors influence the mating system are, at present, unknown.

The hybridogenetic system can apparently allow *R. esculenta* a large degree of "flexibility" in coping with changing habitats through the presence of the host species

R. lessonae. While *R. lessonae* must adapt to newly colonized habitats through processes of natural selection, *R. esculenta* can immediately counter any loss of relative performance by "borrowing" genomes from locally adapted host species through recurrent hybridization (Vrijenhoek et al., 1977; Vrijenhoek, 1989), as well as expressing the *R. ridibunda* genome, which may provide additional adaptive variation. Thus, *R. esculenta* should quickly increase in population size at newly created habitats that are typically of low productivity. The unique feature of the *R. lessonae-R. esculenta* complex is the ability for individuals to reproduce sexually, and alter their fitness by the choice of mates and to "outcross" with locally adapted host species. Studies of this mating system in relation to pond habitats and the relative frequency of hybrids in breeding populations should prove essential to understanding more about the evolution and maintenance of this system.

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