Cohort variation in offspring growth and survival: prenatal and postnatal factors in a late-maturing viviparous snake

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Summary

1. Recruitment to adulthood plays an important role in the population dynamics of late-maturing organisms as it is usually variable. Compared to birds and mammals, few studies assessing the contributions to this variation of environmental factors, offspring traits and maternal traits have been carried out for late-maturing snakes.

2. Cohort variation in recruitment through offspring growth and survival in the meadow viper (*Vipera ursinii ursinii*) was evaluated from 13 years of mark–recapture data collected at Mont Ventoux, France. In this species, females are mature at the age of 4–6 years and adult survival and fecundity rates are high and constant over time.

3. Offspring were difficult to catch during the first 3 years of their lives, but their mean annual probability of survival was reasonably high (0.48 ± 0.11 SE). Mass and body condition at birth (mass residuals) varied significantly between years, decreased with litter size, and increased with maternal length.

4. Cohorts of offspring in better condition at birth grew faster, but offspring growth was not affected by sex, habitat or maternal traits.

5. Survival varied considerably between birth cohorts, some cohorts having a high-survival rate and others having essentially no survivors. No difference in mass or body condition at birth was found between cohorts with 'no survival' and 'good survival'. However, offspring survival in cohorts with good survival was positively correlated with mass at birth and negatively correlated with body condition at birth.

6. Thus, variation in offspring performance was influenced by direct environmental effects on survival and indirect environmental effects on growth, mediated by body condition at birth. Effects of maternal traits were entirely channelled through offspring traits.

Key-words: environmental canalization, maternal effects, squamates, variable environment

Introduction

In late-maturing species with a slow demography, adult survival is generally buffered against temporal fluctuations in environmental conditions, whereas these fluctuations have a much more marked effect on the growth and survival of offspring until maturity (e.g. Saether & Bakke 2000; Gaillard & Yoccoz 2003). This is because natural selection favours mechanisms buffering variation in demographic traits to which fitness is more sensitive, and fitness is more sensitive to adult survival or fecundity than to juvenile survival in species

with a slow demography (e.g. Benton & Grant 1996; Doak *et al.* 2005). Empirical studies of late-maturing birds and mammals have indicated that temporal variation in recruitment between birth years – cohort variation – depends critically on parental effects, offspring characteristics and environmental conditions experienced early in life (e.g. Gaillard, Festa-Bianchet & Yoccoz 1998; Lindström 1999; Gaillard & Yoccoz 2003). The postnatal environment may directly affect growth and survival, through variability in the availability of resources to growing individuals, for example. Variation in offspring traits at birth and early environmental conditions may also have long-lasting effects on life history (e.g. Lindström 1999), and maternal traits, such as body size

or age, may affect offspring growth and survival (maternal effects, Mousseau & Fox 1998). Analyses of cohort effects on offspring growth and survival in the wild require long-term longitudinal data that are rarely available for late-maturing snakes. The availability of such data for these ectothermic vertebrates would make it possible to increase the power of evolutionary comparative analyses, which are currently based on birds and mammals (e.g. Saether & Bakke 2000; Gaillard & Yoccoz 2003).

Temperature, moisture and food availability are important factors affecting life-history variation in snakes (reviewed in Shine 2005, 2003; Seigel & Ford 1987). For example, reproductive frequency increases in response to annual increases in prey density in the water python (Madsen & Shine 1999). Female snakes can also adjust the phenotypes of their offspring to match variation in environmental conditions. The mechanisms underlying this reproductive adjustment include a trade-off between litter size and offspring size (larger clutches are composed of smaller offspring, e.g. Kissner & Weatherhead 2005), adaptive control of the timing of reproduction through thermoregulation behaviour or nest site choice (e.g. Brown & Shine 2005), and an effect of maternal traits, such as body length or body condition, on energy allocation to the offspring (e.g. Gregory & Skebo 1998). A trade-off in maternal energy allocation between offspring mass and condition, on the one hand, and offspring number, on the other, has been suggested in many snake species (King 1993). Effects of the timing of maternal reproduction, maternal body length and maternal body condition have also been found in some taxa (Shine 2003). However, as it is very difficult to find and catch juvenile snakes in natural populations (Parker & Plummer 1987), little is known about the relative contributions of environmental factors, offspring traits and maternal traits to variation in offspring growth and survival. The few field studies conducted to date have generated conflicting results. In some species, juvenile survival is more dependent on the timing of reproduction, habitat quality or climate conditions than on offspring or maternal traits (water python, Madsen & Shine 1998; western rattlesnake, Charland 1989; asp viper, Altwegg *et al.* 2005). In contrast, in other species, offspring size and body condition are good predictors of variation in juvenile survival (northern watersnake, Kissner & Weatherhead 2005; keelback snake, Brown & Shine 2005). There is therefore a need to improve understanding of the factors determining variation in offspring performance.

Viperid snakes from zones with a cold climate are of particular interest in studies aiming to improve our understanding of cohort variation in offspring growth and survival in late-maturing snakes, because these snakes may take up to 10 years to mature, they typically have high adult survival rates and their populations are exposed to significant interannual variation in climate and food conditions (e.g. Parker & Plummer 1987; Lourdais *et al.* 2004; Altwegg *et al.* 2005). The meadow viper *Vipera ursinii ursinii* (Bonaparte, 1835) is a vulnerable European viper species and, as such, has been the focus of a continuous demographic study since 1979 at

Mont Ventoux, in south-eastern France. This population is characterized by female maturity at an age of 4–6 years, biennal reproduction, high annual adult survival rates (*c.* 70–80%), and small litters of large offspring (usually 2–4 offspring weighting about 2–3 g, see Baron *et al.* 1996). A first life-history analysis based on capture–recapture data collected from 1980 to 1994 demonstrated only small differences in adult survival, breeding frequencies, fecundity and litter mass between years (Baron *et al.* 1996). The growth rate of this population is also more sensitive to adult survival than to juvenile survival and reproduction (Ferrière *et al.* 1996), suggesting weak selection against temporal variation in juvenile performance, as reported for other late-maturing vertebrates (Gaillard & Yoccoz 2003).

In 1994, we began to collect longitudinal data on individuals marked at birth. Variation in offspring growth and survival rates was analysed as a function of birth year, offspring traits (mass and body condition at birth) and maternal traits (litter size, parturition date, maternal length and post-partum body condition). These four maternal traits were investigated because other studies of juvenile snakes have reported a trade-off between offspring traits and litter size, and potential effects on offspring fitness of parturition date, maternal body size or maternal body fat reserves (Seigel & Ford 1987; King 1993; Shine 2003). In this study, we address three questions: (i) How do offspring traits vary as a function of maternal traits and year of birth? (ii) Is there significant cohort variation in offspring growth and offspring survival rates? (iii) How do offspring traits, maternal traits and birth year influence variation in offspring growth and survival?

Animals, methods and statistics

STUDY SPECIES AND SITE

The meadow viper inhabits grasslands on calcareous soils at altitudes of 900–2200 m in Western Europe. Mont Ventoux in south-eastern France (altitude of about 1430 m, 44°18′ N, 5°26′ E) hosts a small, isolated population that has been monitored by one of us (J.-P. B.) since 1979. The study area covers 4 ha within a 25-ha zone of favourable habitat and includes a north-facing, largely mesic hillside of habitat that has been modified by human activity (referred to hereafter as habitat A) and a south-facing, more xeric hillside of pristine habitat (habitat B). These two contiguous sites are of similar size and are separated at the bottom by a 10-m-wide asphalt road, strongly limiting dispersal movements; snakes inhabiting either site display differences in terms of life-history traits (Baron *et al.* 1996). Mating occurs during the last 2 weeks of May and gestation extends from late May or early June until mid-August or early September. Females produce a total of two to seven offspring per breeding event and viable neonates (*c.* 90% of the litter) are autonomous at birth (Baron *et al.* 1996). Field observations suggest that juveniles are highly philopatric, remaining within the maternal range during the first years of life (Baron, personal observation).

Meadow vipers of all age and sex classes were sought, captured, and individually marked upon first capture by scale clipping during twice-yearly capture campaigns (each lasting 10–14 days) carried out in May, during the mating season, before ovulation, and in August or September, before parturition. Captured individuals were transferred to the laboratory for measurement of body size (snout–vent length, SVL), to the nearest mm, and body mass, to the nearest 0·1 g. In the summer, the sexual maturity and reproductive status (non-reproductive, pregnant or post-pregnant) of each captured female were assessed visually and by palpation of the abdominal cavity. From 1994 to 2007, gravid females captured before parturition were kept in the laboratory until they gave birth and their healthy neonates were all marked individually by scale clipping ($n = 285$ individuals, 78 litters). Females were kept in individual cages, under optimal thermal gradient conditions (min. = 10 °C, max. = 32 °C), with free access to water. They were fed after parturition. Females were kept for a mean of 13·2 days ($\pm 8·9$ SD) in the laboratory before parturition. Mothers were weighed within 1 day of parturition, their total litter size was counted, and offspring born in the laboratory were measured (SVL and mass) and sexed by counting subcaudal scales (Baron 1997). Mothers were released with offspring at the last capture location, a mean of 11 days ($\pm 6·96$ SD) after parturition. Offspring were subsequently recaptured and measured (SVL and mass) during recapture sessions, thus providing cohort-specific data on offspring growth and survival.

ANALYSIS OF OFFSPRING MASS AND CONDITION AT BIRTH

Offspring mass and condition at birth were examined for annual, spatial (differences between the two habitats) and interindividual variation with mixed-effects models using *lme* (Pinheiro & Bates 2002) in R 2.6.0 (http://cran.r-project.org/). We used data from all cohorts marked and measured at birth ($n = 282$ neonates, 77 litters, 12 birth years). Body condition at birth was defined as the residual of a linear regression of body mass again SVL, obtained by a standard ANCOVA. Body mass was preferred over SVL for analyses of size variation between offspring, because offspring mass can be measured more accurately than SVL, mass provides a better reflection of maternal energy allocation to the offspring than SVL and mass has frequently been used in previous studies of temporal variation in offspring fitness traits in reptiles (e.g. Sinervo 1990). Annual variation was assessed by treating birth year as a random effect and controlling for random variation between litters by including the random effect of litter identity, nested within birth year and habitat effects. The fixed effects of sex, habitat, maternal length (SVL) and post-partum body condition, parturition date and total litter size were also included. Random effects were investigated with likelihood ratio tests, and fixed effects were investigated by *F*-tests based on the usual (REML) conditional estimate of the vari-

ance. A backward procedure was used for model selection, with the removal of non-significant terms ($P > 0·05$).

ANALYSIS OF OFFSPRING GROWTH

A nonlinear von Bertalanffy model was used to analyse individual profiles of change in body size (SVL) before the analysis of offspring growth. This model has three parameters (SVL at birth, exponential growth rate and asymptotic SVL) and was fitted with the *nlme* procedure (Pinheiro & Bates 2002) to individual data collected from birth to the age of at least three or 4 years ($n = 23$ individuals and $N = 116$ observations, including measurements at birth). The model included random individual variation for the three parameters and indicated that the increase in SVL was linear until the age of 4 years (see Results section). All repeated measurements of SVL from birth to the age of 4 years were therefore used to measure growth rate early in life, referred to hereafter as *offspring growth rate*. Only individuals with at least two observations were included in this analysis ($n = 38$ individuals and $N = 109$ observations, including measurements at birth). A mixed-effects model was used to fit a linear growth model assuming random variation between individuals in mean SVL (intercept term) and growth rates. Growth rates were assessed as the slope of the linear regression of SVL against the growth interval. The growth interval was defined as the number of active days since birth date and was calculated, as described by Blouin-Demers, Prior & Weatherhead (2002), as the number of days elapsed between birth date and focal capture date minus the number of days spent in hibernation and emergence, taken to occur from 1 October to 30 April in our study population (Baron *et al.* 1996). Fixed effects of birth year, sex and habitat on growth rates were investigated by including an interaction term between age and each factor in the model. The fixed effects of offspring traits (mass and body condition at birth) and maternal traits (SVL, parturition date, litter size and post-partum body condition) were then included. A backward procedure was used to select the most adequate model.

ANALYSIS OF OFFSPRING SURVIVAL

Effects of cohort, age, sex and habitat on recapture rates and survival rates and effects of offspring traits (mass and body condition at birth) and maternal traits (SVL, parturition date, litter size and post-partum body condition) on survival rates were investigated with capture–mark–recapture (CMR) models in MARK (White & Burnham 1999). Models for open populations, allowing the simultaneous estimation of capture and survival probabilities (Lebreton *et al.* 1992), were used. Juvenile survival (survival during the first year of life) and subadult survival (survival from the age of 1 year to sexual maturity) were assessed with a data set consisting of the yearly recapture histories of offspring marked at birth and recaptured in the field until the age of 5 years, by which time most of the snakes in the study population had reached maturity (Baron *et al.* 1996). General models (listed in

Table 2) used for model selection included only additive effects, because we had no *a priori* expectations concerning potential interactions between sex, habitat, age or cohort effects. Moreover, given the small size of the sample studied, constrained general models excluding interaction terms were used, to avoid the problem of parameter identifiability.

The goodness-of-fit (GOF) of general CMR models was tested with a parametric bootstrapping approach in MARK (White & Burnham 1999). The bootstrap test (1000 simulations) detected slight but significant overdispersion (see Table 2, for more details), possibly due to heterogeneity in capture or survival rates between families. The overdispersion coefficient was estimated as the deviance of the general model divided by the mean deviance of the bootstrap simulations and a quasi-likelihood approach was used for model selection in MARK (Anderson & Burnham 1994). Model selection and hypothesis testing were carried out with the quasi-Akaike Information Criterion, corrected for small sample size (QAICc; Anderson & Burnham 1994). QAICc was obtained by penalizing the deviance of the model with a value proportional to the number of identifiable parameters in the model, thus providing a balance between model parsimony and model fitting (Burnham & Anderson 1998). A symmetric approach was used to test all additive models between the initial model and constant models in a backward selection procedure. The best model was chosen from among those with the lowest QAICc values.

Variation in mean annual survival until maturity between birth cohorts was first analysed, controlling for potential heterogeneity in capture probabilities between cohorts, age classes, sexes and habitats. Seven birth cohorts were considered (1994, 1996–2000 and 2002), totalling 178 individuals (see Table 2a). As described by Lebreton *et al.* (1992), the model best describing variation

in capture probabilities was first selected, followed by the models best describing cohort variation in survival. Based on this analysis, two birth years were identified as essentially 'no survival' cohorts (see Results section). A restricted data set, excluding these cohorts, was therefore used to investigate the effects of age, sex, birth year and habitat on annual survival rates; five birth cohorts were thus analysed, totalling 147 individuals (see Table 2b). The initial model used was the model best describing variation in capture probabilities selected with the full data set (Table 2a). It included additive effects of age, cohort, sex and habitat on survival probability.

In a third model selection procedure, the effects of offspring traits (body mass and body condition at birth) and maternal traits (length, parturition date, post-parturition body condition and litter size) on survival were investigated (see Table 2c). Offspring and maternal traits were standardized before analysis. The initial model was the best model identified in the previous selection procedure (see Table 2b) and included only additive effects.

Results

OFFSPRING TRAITS AT BIRTH

The results of the analysis of annual, spatial, and maternal factors of variation in offspring mass and condition at birth are summarized in Table 1. Both mass and body condition at birth varied significantly between birth years (respectively, $\chi^2_1 = 9.66$, $P = 0.002$ and $\chi^2_1 = 14.61$, $P = 0.0001$). In addition, mass at birth increased with maternal length, decreased with parturition date and total litter size but did not differ between sexes or habitats. Body condition at birth was not influenced by maternal length, but decreased with

Table 1. Effects of maternal traits, habitat and sex on body mass at birth and body condition at birth (mass controlled for offspring length by analysis of covariance) in the meadow viper, *Vipera ursinii ursinii*, at Mont Ventoux, France. Sample size: $n = 282$ neonates, 77 litters, 12 birth years. Results (estimates and marginal test statistics) were obtained by backward elimination of the non-significant effects in mixed-effects models. Model intercepts are given for a mean maternal length (370 mm), litter size (4·09 eggs), and parturition date (7 September), and for the offspring of females from habitat A

	Body mass at birth (g)			Body condition at birth (g)		
	Estimate $\pm$ SE	F-test$_{[nd.f., dd.f.]}$	P-value	Estimate $\pm$ SE	F-test$_{[nd.f., dd.f.]}$	P-value
Fixed effects						
Intercept	2·90 $\pm$ 0·066	–	–	2·97 $\pm$ 0·056	–	–
Maternal length (mm)	0·004 $\pm$ 0·001	14·04$_{[1, 63]}$	0·0004	0·001 $\pm$ 0·0008	1·29$_{[1, 61]}$	0·26
Total litter size	−0·142 $\pm$ 0·036	15·06$_{[1, 63]}$	0·0003	−0·082 $\pm$ 0·021	15·04$_{[1, 62]}$	0·0003
Parturition date (days)	−0·013 $\pm$ 0·005	5·82$_{[1, 63]}$	0·02	−0·009 $\pm$ 0·003	6·58$_{[1, 62]}$	0·01
Habitat (B vs. A)	−0·023 $\pm$ 0·090	0·06$_{[1, 62]}$	0·80	−0·114 $\pm$ 0·046	6·16$_{[1, 62]}$	0·01
Sex (male vs. female)	0·018 $\pm$ 0·037	0·25$_{[1, 197]}$	0·62	0·063 $\pm$ 0·027	5·44$_{[1, 189]}$	0·02
	Estimate (95% CI)			Estimate (95% CI)		
Random effects						
Birth year	0·179 (0·097, 0·329)			0·142 (0·081, 0·249)		
Family identity within year	0·281 (0·224, 0·351)			0·148 (0·112, 0·195)		

Table 2. Mark–recapture models for variation in the probabilities of capture and survival for the offspring of meadow vipers, *Vipera ursinii ursinii*, at Mont Ventoux, France. Three model selection procedures are presented (a–c). Model selection procedure (a) involved the entire data set (seven cohorts). Model selection procedures (b) and (c) involved the restricted data set, with the exclusion of two cohorts having essentially no survivors (see main text and Fig. 2a). QAICc is the quasi-Akaike Information Criterion corrected value. ΔQAICc is the difference in QAICc with the best model. QAICc weight is the model likelihood relative to all models. Model likelihood (*L*) is that relative to the best models. Rank is the number of estimated parameters. At each step, the most informative models according to the QAICc are indicated in bold when ΔQAICc < 2

Model number and name	QAICc	ΔQAICc	QAICc weight	L	Rank	Deviance
(a) Variation in capture probabilities and cohort variation in annual survival probabilities*						
1 Φ_{cohort} P_{a_2class}	**297·646**	**0·000**	**0·711**	**1·000**	**10**	**276·603**
2 Φ_{cohort} P_a	301·663	4·017	0·095	0·134	12	276·170
3 Φ_{ct} P_{a_2class}	301·738	4·092	0·092	0·129	4	293·553
4 Φ_{cohort} P_{a+sex}	303·359	5·713	0·041	0·058	13	275·609
5 Φ_{cohort} P_{a+zone}	303·410	5·764	0·040	0·056	13	275·660
6 Φ_{cohort} $P_{a+zone+sex}$	304·964	7·318	0·018	0·026	14	274·935
7 Φ_{cohort} $P_{cohort+a}$	310·288	12·642	0·001	0·002	18	270·919
8 Φ_{cohort} $P_{cohort+a+zone}$	311·888	14·243	0·001	0·001	19	270·126
9 Φ_{cohort} $P_{cohort+a+sex}$	312·306	14·660	0·000	0·001	19	270·543
10 Φ_{cohort} $P_{cohort+a+zone+sex}$	313·970	16·324	0·000	0·000	20	269·791
11 Φ_{cohort} P_{ct}	329·132	31·486	0·000	0·000	8	312·456
12 Φ_{cohort} $P_{zone+sex}$	332·812	35·166	0·000	0·000	10	311·769
(b) Spatial, sexual and age variation in annual survival probabilities†						
13 Φ_{a1} P_{a_2class}	**294·050**	**0·000**	**0·209**	**1·000**	**5**	**283·726**
14 Φ_{a_linear} P_{a_2class}	**294·113**	**0·063**	**0·203**	**0·969**	**4**	**285·898**
15 $\Phi_{a1+zone}$ P_{a_2class}	**294·352**	**0·302**	**0·180**	**0·860**	**6**	**281·896**
16 Φ_{cte} P_{a_2class}	**295·362**	**1·312**	**0·109**	**0·519**	**4**	**287·147**
17 Φ_{a+sex} P_{a_2class}	296·104	2·054	0·075	0·358	6	283·647
18 Φ_{zone} P_{a_2class}	296·290	2·240	0·068	0·326	5	285·965
19 Φ_a P_{a_2class}	297·311	3·261	0·041	0·196	7	282·699
20 Φ_{a+zone} P_{a_2class}	297·623	3·573	0·035	0·168	8	280·832
21 $\Phi_{zone+sex}$ P_{a_2class}	298·398	4·348	0·024	0·114	6	285·942
22 Φ_{a+sex} P_{a_2class}	299·401	5·351	0·014	0·069	8	282·609
23 $\Phi_{a+zone+sex}$ P_{a_2class}	299·723	5·673	0·012	0·059	9	280·729
24 $\Phi_{cohort+a}$ P_{a_2class}	299·935	5·885	0·011	0·053	11	276·460
25 $\Phi_{cohort+zone+sex}$ P_{a_2class}	300·959	6·909	0·007	0·032	10	279·737
26 $\Phi_{cohort+a+zone}$ P_{a_2class}	301·138	7·088	0·006	0·029	12	275·385
27 $\Phi_{cohort+a+sex}$ P_{a_2class}	301·993	7·943	0·004	0·019	12	276·240
28 $\Phi_{cohort+a+zone+sex}$ P_{a_2class}	303·067	9·017	0·002	0·011	13	275·010
(c) Effects of body mass and body condition on survival probabilities†						
29 $\Phi_{a1+mass1+cond1}$ P_{a_2class}	**286·153**	**0·000**	**0·502**	**1·000**	**7**	**271·541**
30 $\Phi_{a1+mass+cond}$ P_{a_2class}	**286·472**	**0·319**	**0·428**	**0·853**	**7**	**271·860**
31 $\Phi_{a1+mass1}$ P_{a_2class}	291·302	5·149	0·038	0·076	6	278·846
32 $\Phi_{a1+mass}$ P_{a_2class}	293·783	7·629	0·011	0·022	6	281·326
13 Φ_{a1} P_{a_2class}	294·050	7·897	0·010	0·019	5	283·726
33 $\Phi_{a1+cond}$ P_{a_2class}	294·703	8·550	0·007	0·014	6	282·246
34 $\Phi_{a1+cond1}$ P_{a_2class}	295·995	9·842	0·004	0·007	6	283·538

*Bootstrap GOF test based on model 7 ($P = 0\cdot030$, $c = 1\cdot30$). †Bootstrap GOF test based on model 24 ($P = 0\cdot033$, $c = 1\cdot25$). Φ = survival probability, P = capture probability, cohort = birth year, ct = constant, zone = habitat effect, sex = sex effect, a = age effect (a = full age classification, a1 = 1 year old vs. other age classes, a_2class = 1–2 years old vs. 3–4 years old, a_linear = linear age effect), mass = body mass at birth (mass1 = effect limited to juvenile survival), cond = body condition at birth (cond1 = effect limited to juvenile survival).

total litter size and parturition date. Newborn females were leaner than males, and newborns were leaner in habitat B than in habitat A (see Table 1).

OFFSPRING GROWTH

The nonlinear growth curves of 23 vipers recaptured at least once after the age of 4 years showed no difference between the sexes in exponential growth rate or asymptotic body size (all $P > 0\cdot16$). However, random individual variation in body growth rate ($\chi^2_1 = 11\cdot44$, $P = 0\cdot0007$) and asymptotic body size ($\chi^2_1 = 32\cdot61$, $P < 0\cdot0001$) was observed. Initial size, exponential growth rate and asymptotic size were estimated at 138·6 mm (134·7, 142·6) (95% confidence interval CI), 0·38 year^{-1} (0·32, 0·46) and 382·9 mm (367·9, 397·9), respectively. SVL increased almost linearly with age from birth until the age of 4 years (Fig. 1a) and offspring growth rates were calculated for each individual during this linear growth phase. A first analysis of temporal variation in offspring growth revealed significant effects of birth year ($F_{6,64} = 3\cdot21$, $P = 0\cdot008$) but no effects of sex or habitat (all $P > 0\cdot77$). An analysis of prenatal and postnatal factors
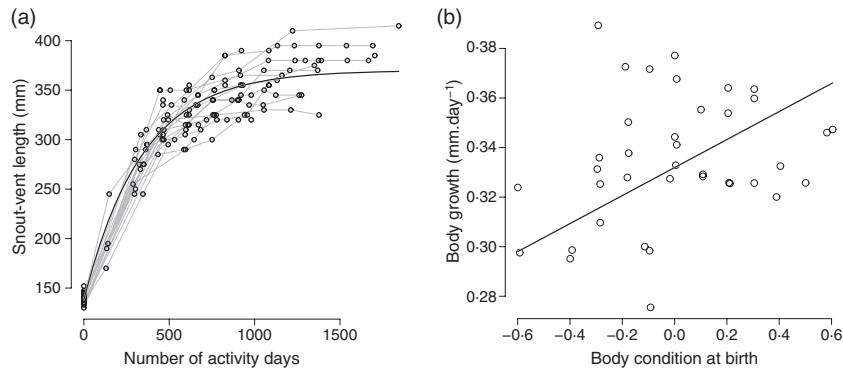
Fig. 1. Offspring growth in the meadow viper. (a) Snout–vent length (SVL) with respect to number of days of activity since birth (i.e. excluding hibernation and emergence periods). Body growth was almost linear until maturity (*c.* 700 active days). Regression line corresponds to the mean von Bertalanffy growth curve and growth trajectories of individuals with more than three measurements are drawn. (b) Regression between offspring growth rate and body condition at birth. Daily increases in SVL were estimated during the linear growth phase, from birth to the age of 4 years, for 38 snakes marked at birth.

affecting the variation in growth showed that offspring growth rates increased with body condition at birth ($F_{1,69} = 6.06$, $P = 0.02$), but did not vary with mass at birth or maternal traits (all $P > 0.05$). Neonates in good condition grew faster early in life (Fig. 1b). The year of birth no longer had a detectable effect on growth rates if the analysis was controlled for body condition ($F_{6,63} = 1.44$, $P = 0.21$).

OFFSPRING SURVIVAL

Annual survival probabilities of offspring were estimated with mark–recapture data for seven birth cohorts. The various models compared are summarized in Table 2a. The prob-

ability of capture did not vary between cohorts, sexes or habitats, but did vary between age classes. Capture probabilities were lower in snakes aged 1–2 years [0.13 (0.07, 0.21) 95% CI] than in snakes over the age of 2 years [0.56 (0.38, 0.73), estimates from model 1 in Table 2a]. Taking this finding into account, mean annual survival to the age of 5 years differed markedly between birth cohorts (Table 2a, model 1 vs. model 3: $\chi^2_6 = 16.95$, $P = 0.009$) and this variation was mainly due to two cohorts having no survivors (1997 and 1999, Fig. 2a). The mean annual probability of survival was 0.48 (± 0.11 SE). For two cohorts (1997 and 1999) essentially no juveniles survived, whereas the other five cohorts displayed higher, constant survival rates. A variance compo-
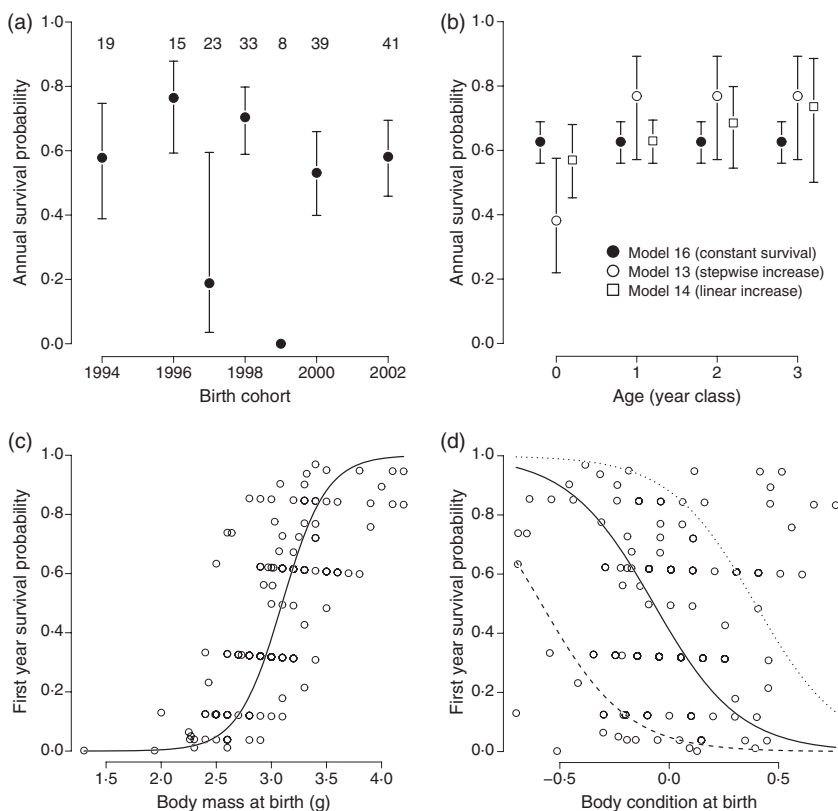

Fig. 2. Offspring survival in the meadow viper. (a) Mean (95% CI) annual survival for each birth cohort (sample size given for each cohort) from birth until the age of 5 years, according to model 1 in Table 2a. (b) Mean annual survival per age class after exclusion of the 1997 and 1999 birth cohorts. The best mark–recapture models predict a stepwise increase, a linear increase, or constant survival with age (according to models 13, 14 and 16, respectively, in Table 2b). Predicted juvenile survival probabilities (survival from birth to 1 year) plotted against mass at birth (c) and body condition at birth (d) according to model 29 in Table 2c. For mass at birth, the regression line is drawn for the mean value of body condition at birth for the population. For body condition at birth, regression lines are drawn for three values of mass at birth (2.5 g, dashed; 3.0 g, solid; 3.5 g, dotted).

nents approach provided a measurement of annual variance in survival probabilities of $\sigma^2 = 0.073$, with a 95% CI of (0.028, 0.37).

The two birth cohorts without survivors were excluded and the effects of age, sex, birth year and habitat on annual survival probability were investigated. In this data set, no difference in survival was detected between the sexes (Table 2b) or between the cohorts [LRT test of cohort effect: $\chi^2_4 = 6.24$, $P = 0.18$; mean annual survival probability = 0.63 (0.56, 0.69) 95% CI], but survival probabilities tended to increase with age (Fig. 2b). However, the Akaike information-based criterion could not discriminate between a linear increase in survival with age (LRT test of the linear effect: $\chi^2_1 = 2.17$, $P = 0.14$), a stepwise increase in survival after the first year of life (LRT test of the stepwise increase: $\chi^2_1 = 3.42$, $P = 0.06$) and constant survival rates with age (all the models shown in bold in Table 2b are within ± 2 QA-ICc scores of the best model). There was some support for variation in survival between habitats, based on QAICc, with slightly higher levels of survival in habitat B (logit contrast = 0.46 ± 0.35 SE), but this spatial difference was not significant ($\chi^2_1 = 1.80$, $P = 0.18$).

Comparisons of birth cohorts with high levels of survival and no survival revealed no significant difference in mass or condition at birth (mass: $F_{1,5} = 0.43$, $P = 0.54$; condition: $F_{1,5} = 0.06$, $P = 0.82$). Within the birth cohorts with high-survival rates (five cohorts), maternal traits had no significant effect on offspring survival (see Table S1). However, effects of mass at birth ($\chi^2_1 = 12.00$, $P = 0.0005$) and body condition at birth ($\chi^2_1 = 7.31$, $P = 0.007$) were significant (see Table 2c). Janzen & Stern's formula (1998) for standardized selection gradients (β) indicated strong directional selection on mass at birth, with heavier offspring having higher survival rates (Fig. 2c, model 29: $\beta = 2.61$, model 31: $\beta = 0.89$), and, when controlling for the effects on offspring survival of mass at birth, significant negative directional selection on body condition at birth (Fig. 2d, model 29: $\beta = -1.56$). The best model (model 29) indicated that these effects of offspring mass at birth and offspring condition at birth were restricted to juvenile survival (survival from birth to the age of 1 year). However, this best model could not be distinguished from the second-best model (model 30) including effects of offspring traits on mean offspring survival from birth to the age of 4 years (see Table 2c).

Discussion

Studies of late-maturing snakes have had limited success in documenting and explaining how environmental factors, offspring traits, and maternal traits influence variation in offspring growth and survival and, therefore, recruitment to adulthood (Shine 2003; Brown & Shine 2005). In the late-maturing meadow viper, variation in offspring mass and condition at birth is consistent with trade-offs in energy allocation between offspring size and number. There is also substantial cohort variation in offspring size, growth and survival. Cohort variation in offspring growth results from vari-

ation in body condition at birth: higher body condition at birth was found to be correlated with faster growth. The mean annual probability of survival from birth to maturity varied considerably between birth cohorts, some of which displayed high-survival rates, whereas others displayed no survival. For individuals from high-survival cohorts, offspring survival increased with mass at birth, but decreased with body condition at birth, particularly during the first year of life. Thus, the maternal traits investigated here had no direct effects on offspring growth and survival, but did have some indirect effects mediated by variation in offspring traits.

DETERMINANTS OF OFFSPRING SIZE

The variation in offspring size at birth (mass and condition) reported here is consistent with the results of previous studies of maternal effects in snakes (reviewed in Shine 2003). The relationship between offspring size and fecundity can be interpreted as a trade-off in energy allocation between offspring size and offspring number, as found in other late-maturing snakes (e.g. King 1993; Kissner & Weatherhead 2005). In viviparous snakes, this trade-off may be mediated by maternal provisioning of eggs during vitellogenesis, as shown for lizards (Troyer 1987; Sinervo 1990 and references therein). Offspring mass was also constrained by maternal body length and offspring condition was lower in litters born later in the season. A similar positive effect of maternal body length has been reported in some late-maturing snakes when the energy available for reproduction is not limiting and is due to the constraints imposed on litter volume by maternal body volume (Lourdais *et al*. 2002).

VARIATION IN OFFSPRING GROWTH AND SURVIVAL BETWEEN COHORTS

The meadow viper grows rapidly, and almost linearly, early in life, but its growth slows when it reaches maturity. No sexual dimorphism or spatial variation in size or growth rate was detected, and our analysis highlighted the value of a single individual covariate, body condition at birth, as a predictor of offspring growth rates. Birth year was a significant determinant of body condition at birth and, although offspring growth varied significantly between birth years in a univariate analysis, no such variation was observed if the analysis was controlled for body condition at birth. Thus, annual variation in body condition at birth resulted in cohort variation in body growth. Body condition at birth may reflect offspring body fat reserves and the contribution of post-hatching yolk (Charland 1989; Jayne & Bennett 1990). Meadow vipers do not feed until summer, when preys become available. Juveniles must therefore rely on their reserves during the first few months of activity and initial fat stores may be crucial for early growth (Baron 1997). Furthermore, as there is a trade-off between offspring condition at birth and offspring number (see above), we hypothesize that there is also a trade-off between offspring number and offspring

growth, which has not been considered in previous studies of this type in snakes (Niewiarowski & Dunham 1994; Shine 2003). In the meadow viper, faster growth early in life may enable the offspring to reach maturity more rapidly or to reproduce when they attain a larger body size, with potential fitness benefits in terms of reproductive output (Baron *et al.* 1996).

The mark–recapture data were also used to test for factors associated with variation in offspring survival, despite the problems of low recapture rates for immature snakes. A catastrophic collapse of offspring survival was observed in two of the seven birth cohorts monitored. This suggests that young animals in these cohorts were exposed to factors associated with high mortality rates that had little effect on older offspring from previous cohorts. Cohort-specific mortality factors in the meadow viper may include (i) poor maternal investment in offspring, (ii) harsh climatic conditions after release, (iii) unusually high levels of inbreeding or genetic defects, or (iv) inappropriate procedures during maintenance in the laboratory or release (Diller & Wallace 2002; Altwegg *et al.* 2005; Brown, Kéry & Hines 2007). Only one adult born from the cohorts with essentially no survival was found in the population in the field (one field-born animal from 1997). Mortality rates were thus high in both laboratory- and field-born offspring and were therefore not a result of our protocols. No data were available concerning inbreeding or genetic defects, but these defects should be associated with reproductive failures or birth deformities (Ujvari *et al.* 2002). No such abnormalities were observed here. Moreover, there was no significant difference between high- and no survival birth cohorts in terms of offspring mass or body condition at birth. Cold winters have been associated with poor juvenile survival in another European viper (Altwegg *et al.* 2005), but the no survival birth cohorts in this study were not subjected to colder winters (mean winter temperature, 0·43 °C ± 3·85 SD vs. 0·02 °C ± 3·92 SD in cohorts with good survival). Thus, neither annual variation in offspring traits nor differences in mean winter temperature can account for the major differences in offspring survival observed. Instead, we speculate that unusual, short-term winter climate conditions may expose juveniles to a high risk of death. Longer term mark–recapture studies and more detailed meteorological data for the winter would be required to test this hypothesis.

In the two cohorts with essentially no survival, unknown mortality factors swamped the potential expression of differential selection on offspring traits. However, in the five cohorts with higher and more constant levels of offspring survival, three interesting results emerged from analyses of offspring survival as a function of age, habitat, offspring traits and maternal traits. First, mass at birth was a primary determinant of offspring survival. Mass at birth tended to have a stronger effect on survival in the first year of life and none of the maternal traits had a direct effect on offspring survival. Secondly, offspring survival did not differ between sexes, consistent with the data from other late-maturing viperid snakes (Charland 1989; Diller & Wallace 2002; Brown *et al.* 2007). Thirdly, survival tended to increase with age and be higher in habitat, although the power of these statistical tests was too low to resolve the possible effects of age and habitat on survival, and additional data will be required to clarify these patterns.

The influence of mass at birth on offspring survival highlights the importance of 'early start' conditions for offspring performance. Only a few field studies have considered natural selection on offspring size (or mass) at birth in snakes. Also, previous studies of size-dependent selection in wild free-ranging snakes have made use of return rates rather than survival probabilities (but see Kissner & Weatherhead 2005 for an enclosure study). These studies have generated conflicting results, with some species showing evidence for positive selection on offspring size (e.g. Brown & Shine 2005; Kissner & Weatherhead 2005) and others reporting no such selection (e.g. Charland 1989; Madsen & Shine 1998). Directional selection on mass at birth, as estimated by standardized selection gradients, was very strong in birth cohorts with high-survival rates in this study, indicating that meadow vipers produce a small number of large offspring for which maternal investment is critical. Larger young may be born with greater total energy reserves, making them less sensitive to food shortage early in life (e.g. Charland 1989; Brown & Shine 2005). They may also have greater locomotor or foraging capacities (e.g. Jayne & Bennett 1990; Kissner & Weatherhead 2005).

EFFECTS OF BODY CONDITION AT BIRTH ON OFFSPRING GROWTH AND SURVIVAL

Body condition differed significantly between birth years and was negatively correlated with offspring survival. Selection against high body condition at birth seems at odds with the notion that body condition is an indicator of body fat reserves and, thus, of the capacity of offspring to fulfil their metabolic needs early in life. However, the faster growth associated with higher body condition at birth may be associated with mortality costs. In addition to the growth-survival resource allocation trade-off, resource acquisition and foraging activities may expose juvenile snakes to factors associated with a high rate of mortality, such as predators (Forsman, 1993). Moreover, the negative effect of body condition on survival was only apparent if the analysis was controlled for the positive effect of mass (see Table 2c): survival was higher in neonates with a lower body condition (body mass residuals) than in neonates with a higher body condition but the same body mass. Thus, survival rates were higher for longer individuals with a given mass. Jayne & Bennett (1990) put forward several advantages of this 'leanness' to account for viability selection against high body condition values in yearling garter snakes. In the meadow viper, leaner juveniles may have access to narrower and deeper crevices in the soil, and may therefore hibernate in shelters of higher quality, potentially accounting for the higher survival rates of leaner neonates at 1 year.

Body condition had a positive effect on offspring growth but a negative effect on offspring survival, and we could infer

from this that body condition appears to drive a negative correlation between growth and survival. Theoretical analyses have suggested that such a negative correlation may oppose the environmental canalization of body condition at birth, or even favour the evolution of higher variability (discussed in Doak *et al.* 2005). The additive and opposite effects on offspring survival of mass and body condition at birth, and the positive effect of body condition at birth on offspring growth also raise the interesting possibility that selection may favour growth over survival in birth years in which offspring mass is low, and survival over growth in birth years in which offspring mass is high. In birth years with a low offspring mass, offspring survival is low and the production of short but fat, rapidly growing neonates may constitute a 'best-of-a-bad-job' strategy, minimizing losses. In birth years in which offspring mass is high, the offspring will grow to be large even if growth is slow. The production of long, lean neonates with a high likelihood of survival may maximize fitness benefits in these conditions. According to this hypothesis, there should be a negative correlation between body mass and body condition at birth, but no such correlation was detected in a simple regression analysis of our data across birth years ($n = 18$, Pearson's product moment correlation, $r = 0·08$, $P = 0·73$) or across families ($n = 103$, $r = 0·07$, $P = 0·48$). Nonetheless, our adaptive scenario has implications for recent theories about the potential fitness benefits of life-history trait fluctuations and should thus encourage further empirical investigation of environmental canalization in this and other taxa.

Acknowledgements

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Supporting Information

Additional Supporting Information may be found in the online version of this article.

Table S1. Summary of mark–recapture models used to assess maternal effects on juvenile survival

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