



RESEARCH ARTICLE

Are prenatal maternal resources more important in competitive than in benign postnatal environments?

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ABSTRACT

According to theoretical models, the optimal solution of the life-history trade-off between the number and size of offspring depends on the quality of the environment. Offspring size should be more important for their fitness in more competitive environments. This idea was rarely experimentally tested in taxa with prolonged periods of parental postnatal care, such as in birds. Here we manipulated the offspring rearing environment by enlarging or reducing brood size. Enlarged broods suffered greater mortality rates and raised smaller fledglings. Egg size had a significant positive effect on fledging mass and length of tarsus and a nonsignificant effect on wing length. These effects were similar in enlarged-sized as well as reduced-sized broods. We only found a tendency for the predicted interaction between treatment and egg size in the case of nestling mass where egg size had a positive effect in enlarged broods but none in reduced broods. In contrast, in one year we found an opposite interaction where egg size positively affected offspring survival only in reduced broods. More studies that manipulate the offspring rearing environment and follow offspring over the long term are needed to draw general conclusions about context-dependence of early maternal effects.

Keywords: context-dependence, egg size, environmental quality, parental investment

¿Son los recursos maternos prenatales más importantes en ambientes postnatales competitivos o benignos?

RESUMEN

De acuerdo a los modelos teóricos, la solución óptima en la historia de vida entre el número y el tamaño de los hijos depende de la calidad del ambiente. El tamaño de los hijos debería ser más importante en términos de éxito reproductivo en los ambientes más competitivos. Esta idea ha sido evaluada pocas veces experimentalmente en taxa con períodos prolongados de cuidado parental postnatal, como en las aves. Aquí manipulamos el ambiente de cría agregando o reduciendo el tamaño de la nidada. Las nidadas con crías agregadas presentaron tasas de mortalidad mayores y criaron volantones más pequeños. El tamaño del huevo tuvo efectos positivos significativos en el peso y en el largo del tarso de los pichones y tuvo un efecto no significativo en el largo del ala. Estos efectos fueron similares en las nidadas con crías agregadas reducidas de tamaño. Solo encontramos una tendencia en la interacción que predijimos entre el tratamiento y el tamaño del huevo en el caso del peso del pichón, donde el tamaño del huevo tuvo un efecto positivo en las nidadas con crías agregadas pero no en las nidadas reducidas de tamaño. Por el contrario, en un año encontramos una interacción opuesta donde el tamaño del huevo afectó positivamente la supervivencia de los pichones solo en las nidadas reducidas de tamaño. Se necesitan más estudios que manipulen el ambiente de cría de los pichones y que sigan a los pichones al largo plazo para obtener conclusiones generales sobre la dependencia del contexto de los efectos maternos tempranos.

Palabras clave: calidad ambiental, dependencia del contexto, inversión parental, tamaño del huevo.

INTRODUCTION

Parents often face the dilemma whether to invest in a limited manner to many offspring or invest heavily in a smaller number. This trade-off between offspring quality and quantity was first modeled by Smith and Fretwell (1974), who assumed diminishing fitness returns from investment in each offspring. Consequently, every offspring should only receive an intermediate amount of parental resources, and the remaining resources may be

better used for nourishment of further offspring or for parental maintenance. This early model predicted one optimal offspring size both within and between families.

Subsequent models added more complexity and realism to this theory, assuming that fitness returns from parental investment may depend on environmental context. In theory, the initial maternal investment should be more important for offspring performance if they live in harsh and competitive conditions than in mild ones where all offspring have a good chance to survive (Brockelman 1975, Parker and

Begon 1986, McGinley et al. 1987, Fischer et al. 2011, Kuijper and Johnstone 2013). Consequently, harsher habitats should select for large offspring whereas more benign habitats select for small offspring. This spatial and temporal variance in selection pressures is likely the underlying cause of observed egg-size variation between and within species (Fox and Czesak 2000). Recently, this hypothesis was supported in several fish species (Einum and Fleming 1999, Bashey 2008, Rollinson and Hutchings 2013, Riesch et al. 2014).

Studying these ideas in other taxa could test their generality across different life-histories. For example, birds usually have a more-developed parental care in the form of incubation and offspring provisioning, which impacts offspring phenotypes (DuRant et al. 2013, Bowers et al. 2014) and thus could mitigate earlier maternal effects. Therefore, whether context-dependence may induce the same inter-population variability of egg size in birds as was found in fish is unclear. Many avian studies have focused on the relationship between egg size and offspring quality, yet only a few tested if egg-size effects depend on rearing conditions (Krist 2011). We know of only 3 studies that manipulated chick postnatal conditions to test for context-dependence of egg-size effects. In the first, food was supplemented to mimic a mild environment, but egg-size effects were not smaller than in natural nests (Styrsky et al. 2000). Similarly, egg-size effects were not larger in highly competitive enlarged broods (Bonisoli-Alquati et al. 2008); however, they were more pronounced in a stressful condition created by handicapping the parents (Love and Williams 2011). In contrast, between-study comparisons revealed a larger egg-size effect in captivity than in the wild, despite wild conditions likely representing a more stressful environment (Krist 2011). From these few and contradictory results, no general conclusion about context-dependence of maternal effects can be derived, and further studies are therefore warranted.

In this study we experimentally tested the assumption of adaptive scenario for between-population variation in egg size, specifically whether egg size has stronger effect on offspring performance in poor compared to good rearing environments. We created these good and poor rearing environments by experimental manipulation of brood size in the Collared Flycatcher (*Ficedula albicollis*). Enlarged broods represented poor rearing environments as a result of increased competition between nest mates (see also Forbes 2011). Despite poor offspring performance in enlarged broods, we did not find consistent evidence for stronger egg-size effects on nestling fitness-related traits in these competitive conditions.

METHODS

Study Area and Field Experiments

This study was conducted on Velký Kosíř (49°32'N, 17°04'E, 300–400 m a.s.l.), Czech Republic, in 2011, 2012, and 2014. In the breeding season, nest-boxes were monitored for

newly laid eggs of Collared Flycatchers. All eggs were measured for width and length by digital calliper to the nearest 0.01 mm, and egg volume was calculated according to Hoyt's (1979) formula: $\text{volume} = 0.51 \times \text{length} \times \text{breadth}^2$. In altricial birds, sibling competition is a major force that affects the quality of both objective and effective rearing environment (Forbes 2011); therefore, we manipulated brood size to decrease sibling competition in reduced broods and enhance it in enlarged ones. The details of how brood size was manipulated differed between years.

In 2011, complete clutches were exchanged between pairs of nests differing by one in clutch size and matched ($n = 46$) or differing by one ($n = 22$) in laying date of the last egg. Cross-fostering was usually done 2–3 days after the laying of the last egg (mean = 3.13, SD = 1.87, range 1–12). In 2012, 2-day-old chicks were exchanged between nests matched by hatching date and clutch size ($n = 56$) or differing in clutch size by 1 ($n = 12$) or 2 ($n = 2$) egg. In enlarged broods, 2 randomly chosen chicks or eggs were left and the remainder were moved to reduced broods. All young and eggs from reduced broods were fostered in enlarged broods. In cases where clutch sizes were not matched, we left a number of young in enlarged nests, which increased their brood size by 2 young or eggs. In 2014 one of the core young from a reduced brood was taken and transferred to an enlarged brood 2 days after hatching. We transferred only core young (i.e. those that did not suffer from late hatching; see Forbes 2011) to enhance the chance of their survival and thus prolonged sibling competition in enlarged broods. Nests in experimental dyads were matched by hatching date ($n = 88$) or differed by 1 day ($n = 16$). In dyads matched by hatching date, treatment was allocated randomly within dyads; in dyads differing by 1 day, the chick was always transferred from the older to the younger brood to enhance the chance of its survival among younger competitors. The 2 chicks from the original nest (in 2012) or the 1 from the foster nest (in 2014) were marked by trimming part of the down feathers on their heads.

On day 6, nestlings were ringed and weighed. We also noted which nestlings had trimmed feathers. On day 13, nestlings were reweighed and their tarsi (to 0.01 mm) and wings (to 1 mm) were measured. Thereafter, nests were checked at weekly intervals to record the fate of individual nestlings (fledged or died).

Statistical Analyses

The main predictors in all initial statistical models were mean volume of eggs in the nest, treatment (enlarged or reduced), and their interaction. We also included a 3-way interaction between treatment, egg size, and year to check whether potential context-dependence of egg-size effects was apparent only in some years, for example due to difference in our experimental designs. If these interactions

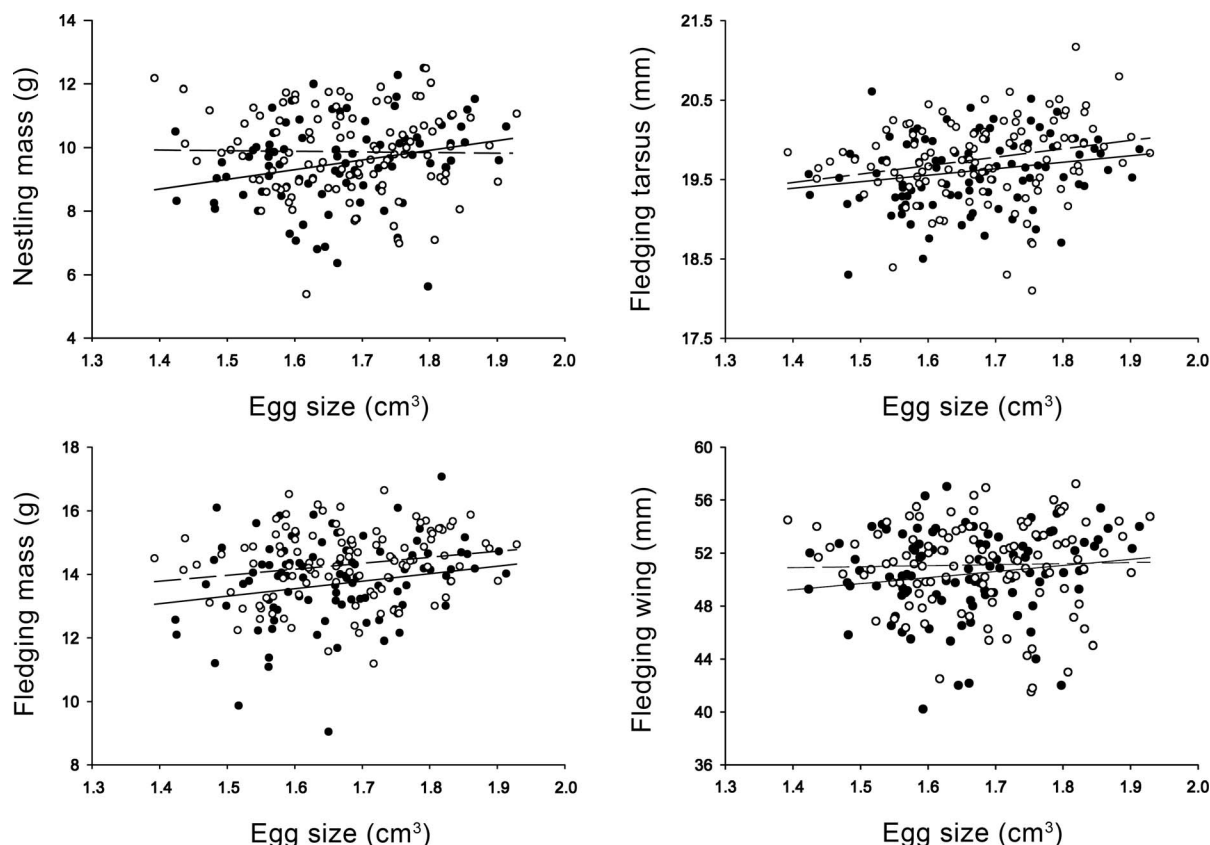


FIGURE 1. Scatterplots showing relationships between egg size and nestlings morphological traits in enlarged (solid circles and lines) and reduced (open circles and dashed lines) broods. Fitted lines are predicted from models that included interactions between treatment and egg size.

were not significant ($P > 0.05$), they were omitted from the final models, starting with the 3-way interaction. Further fixed predictors were year, fitted as a categorical factor, to control for annual effects; laying date to control for seasonal effects; and initial clutch size (see Hauber 2002 for a discussion of potential clutch-size effects). We also included identity of recipient and donor nest-boxes as random factors because some were used in several years.

Response variables were average nestling traits: body mass, length of wing and tarsus, and fledging success. These variables were averaged for focal young only; we omitted from this average the young that were left in original nests (in 2012) or that were transferred to foster nests (in 2014). Thus, mean egg size predicted traits of nestlings that were either cross-fostered (in 2011–2012) or raised in original nests (in 2014). All averages were based solely on fledged young and fledging success solely on broods from which at least one young fledged. We excluded dead chicks from computation of averages to avoid bias due to inclusion of starved nestlings that often have outlying values of morphological traits. We did not include total failures when testing for fledging success because such failures are unlikely to be caused by size of

eggs (Krist 2011); more likely reasons are predation of adults or nest abandonment. All tests were done in SAS, v9.2, Proc Glimmix. We used identity link when response variables were normally distributed (morphological traits) and logit link and event/trial syntax for binomially distributed fledging success.

In theory, egg-size effect can be inflated if eggs are not cross-fostered due to the co-variation of egg size and parental quality (Magrath 1992, Krist and Remeš 2004). In reality, however, this inflation is usually small at best, as demonstrated by a recent meta-analysis (Krist 2011) and a previous study in this population (Krist 2009). Moreover, in this study we are interested in the difference of egg-size effects between treatments, not in absolute egg-size effect within a treatment. Thus our data, despite being based on a mix of nestlings raised in original and foster nests, are well suited for testing the interaction between egg size and quality of the post-natal environment.

RESULTS

Main results are presented as direction and size of egg effects and treatment effects (Figures 1, 2) and statistical

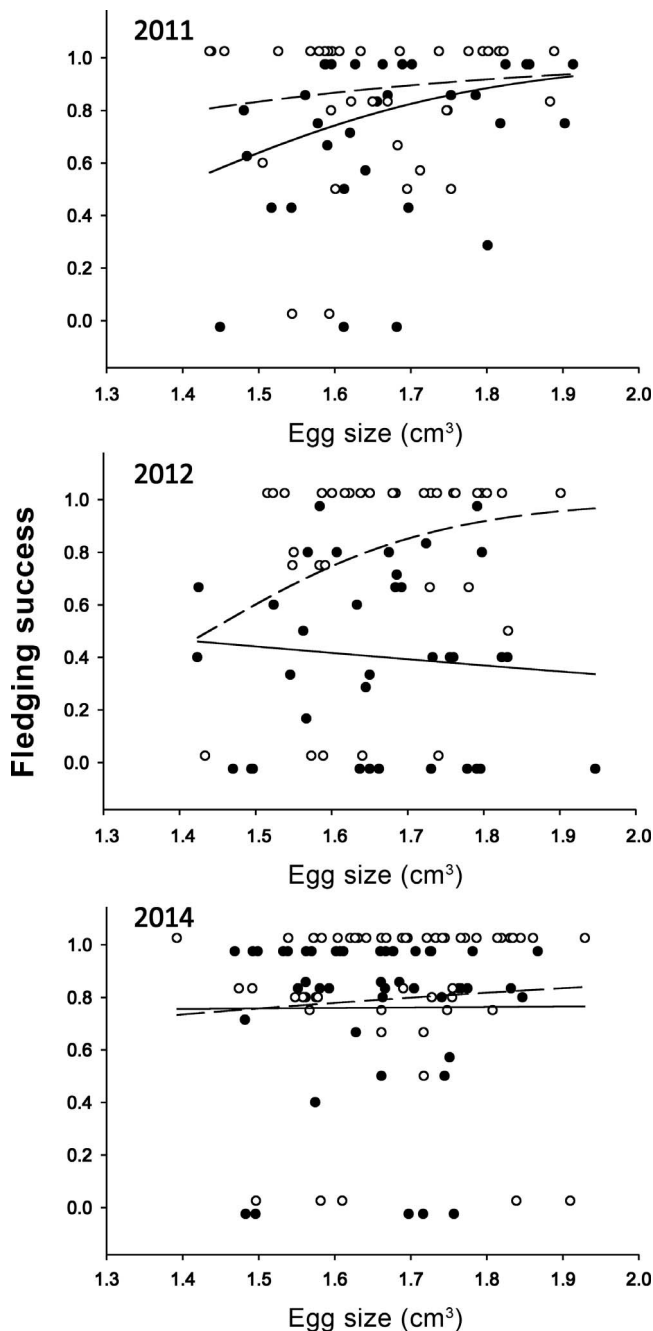


FIGURE 2. Relationship between egg size and fledging success in 3 years of the study. Broods with complete failure are also displayed, but these nests were not used for statistical tests. For nests with complete failure or success, points are slightly shifted above (reduced broods) or below (enlarged broods) from 0 and 1 to make the pattern more visible. Fitted lines are predicted from models that included interactions between treatment and egg size.

significance of these relationships (Table 1). Scatterplots show raw data; however, fitted lines were predicted from models that included the interaction between treatment and egg size. Thus, these predicted values were controlled for effects of other variables in the model. Difference in slopes of these lines would suggest different effect of egg size on offspring performance in enlarged compared to reduced broods (i.e. significant interaction between treatment and egg size). Different height of these lines would suggest treatment effect.

Because the lines are generally lower in enlarged broods, nestlings in these broods were smaller (Table 1; Figure 1) and had a lower rate of survival (Table 1; Figure 2) than those in reduced broods, indicating that our experiment created nest environments with low and high intensity of sibling competition. For example, nestlings in enlarged broods fledged at about 0.5 g lower mass than those in reduced broods, an effect independent of egg mass (Figure 1). Despite lower survival rate, final number of all fledglings was still higher in enlarged broods (total failures excluded; mean \pm SE: 5.22 ± 0.14 vs. 4.03 ± 0.14 , $F_{1, 209} = 37.00$, $P < 0.001$), which holds also for number of focal fledglings (4.56 ± 0.14 vs. 4.03 ± 0.13 , $F_{1, 205} = 7.39$, $P = 0.007$).

Egg size generally had positive effects on all offspring traits (Figures 1, 2), although this effect was not statistically significant for wing length (Table 1). Egg-size effects were not confounded by difference in initial clutch sizes because these 2 variables were not correlated ($r = -0.060$, $n = 242$, $P = 0.354$). We found no consistent evidence that egg-size effects are more important in competitive than in benign postnatal environments. There was only a tendency for stronger egg-size effect on nestling mass in enlarged broods (Figure 1, test of the interaction: $F_{1, 189} = 3.76$, $P = 0.054$). On the contrary, fledging success was more dependent on egg size in reduced broods in 2012, while no difference between slopes was found in 2011 and 2014 (Table 1; Figure 2). Similarly, in the case of other morphological traits, the slope of egg-size effect in reduced broods was similar to that in enlarged broods (Figure 1).

DISCUSSION

Egg size had positive effects on the offspring's fitness-related traits, indicating that parents did not fully compensate for small eggs with more intensive prenatal (incubation behaviour; DuRant et al. 2013) or postnatal (offspring provisioning; Bowers et al. 2014) care. Avian egg size thus might be as important for offspring fitness as in taxa with little parental care, such as in fish. However, unlike in fish, we did not find consistent evidence for context-dependence of egg-size effects. Although our brood size manipulation was effective in creating harsh and mild rearing environments, the slopes relating

TABLE 1. Effects of fixed factors on offspring traits. Only final models after elimination of nonsignificant interactions are shown. DDF = denominator degrees of freedom, *F* = *F*-value, and *P* = *P*-value. Statistical directions of treatment and egg-size effects are given in Figures 1 and 2. Year was fitted as a categorical variable.

	Body mass at 6 days			Body mass at 13 days			Tarsus length			Wing length			Fledging success		
	DDF	<i>F</i>	<i>P</i>	DDF	<i>F</i>	<i>P</i>	DDF	<i>F</i>	<i>P</i>	DDF	<i>F</i>	<i>P</i>	DDF	<i>F</i>	<i>P</i>
Treatment	188	3.55	0.061	198	12.63	<0.001	196	4.12	0.044	194	2.13	0.146	194	0.18	0.672
Egg size	189	2.41	0.122	198	8.61	0.004	197	10.74	0.001	192	1.68	0.196	194	1.13	0.230
Egg size * Treatment													194	0.01	0.940
Egg size * Treatment * Year													194	2.51	0.043
Year	168	2.55	0.081	198	9.34	<0.001	182	9.42	<0.001	176	8.97	<0.001	194	0.37	0.691
Laying date	190	17.58	<0.001	198	9.43	0.002	197	10.85	0.001	191	29.8	<0.001	194	8.83	0.003
Clutch size	189	2.53	0.113	198	5.53	0.020	198	0.16	0.693	192	6.07	0.015	194	5.42	0.021

offspring performance to egg size were generally similar across environments. We found only a tendency for steeper slope in enlarged broods for nestling mass but an opposite pattern (steeper slope in reduced broods) for fledging success in 1 of the 3 study years.

Egg size is often variable between populations (Fox and Czesak 2000, Martin et al. 2006, Marshall and Keough 2008). In the prevailing view, this variation should be adaptive because harsh environments select for large eggs, while in benign habitats egg size is of less importance for offspring survival. Consequently, population differences in egg size could arise either as evolved fixed strategies (Fox and Czesak 2000) or as plastic responses of individuals able to anticipate the quality of the offspring environment and adjust their eggs accordingly (e.g., Garant et al. 2007). However, experiments that manipulate the offspring-rearing environment to test its interaction with egg size on offspring and maternal fitness are rare in taxa in general (Fox and Czesak 2000) and in birds in particular (Krist 2011, Williams 2012). Thus, an alternative explanation could be that inter-population variation is largely non-adaptive and has arisen due to proximate constraints such as temperature, precipitation, and food supply at the time of egg formation. The results of our study somewhat support this nonadaptive scenario because we did not find consistent evidence for stronger egg-size effects in poor rearing environments. However, several potential issues should be considered.

First, we tested egg-size effects on traits that are correlates of offspring fitness, not on true offspring fitness itself. This major weakness is unfortunately a rule rather than an exception in this field (Krist 2011) and in other fields (Hunt et al. 2004). Fledging traits are strongly correlated with first year survival (Lindén et al. 1992, Kruuk et al. 2001); however, recently it was shown that a large part of this correlation might be caused by annual effects, not causal effects of fledgling traits (Bouwhuis et al. 2015). So the challenge for future work is to follow

offspring over the long-term, ideally through their lifetime, which is inevitably a difficult task.

Second, we experimentally created environments of 2 qualities and looked at the difference in egg-size effects between them. More elegant evidence for context-dependence would be the gradual change of egg-size effects across an environmental gradient (Rollinson and Hutchings 2013); however, such studies are usually observational because experimentally inducing environmental gradients would be challenging. Nevertheless, this experimental approach has the strength of decoupling the potential correlations between the factor of interest, here intensity of sibling competition, and other unmeasured variables.

Third, here we studied egg-size effects between broods. Although egg-size variability is usually much greater between females rather than within them (Christians 2002), which makes tests conducted on the former level more powerful (Krist 2011), nestlings from different broods do not directly compete, which might decrease the chance to reveal context-dependence of egg-size effects between nests. It is difficult to predict which of the 2 mechanisms will prevail, and thus the level of context-dependence should be easier to find. In a previous study we generated extra variation of egg size within clutches and found evidence for context-dependence of egg-size effects within broods (Krist and Munclinger personal communication). Another possibility to test environmental specificity of egg-size effects would be to compare offspring of different hatching orders. In species with substantial hatching asynchrony, marginal offspring that hatched late live in an effectively more competitive environment than core offspring (Forbes 2011). Thus, egg size should be more important for fitness of marginal than core offspring.

We concentrated our discussion on only one form of maternal effects: egg size. It is clear that there are many other types of prenatal effects, such as quality of

incubation (DuRant et al. 2013) and amount of androgens (Gil 2008), carotenoids (Saino et al. 2003), immune factors (Saino et al. 2002), and vitamins (Matrková and Remeš 2014). All of these prenatal as well as the postnatal effects, such as nestling provisioning, may also have context-specific effects on offspring fitness; however, this possibility is even less explored than egg-size effects. Clearly more data are needed to draw more general conclusions about context-specific effect of parental investment on offspring fitness.

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Ethics statement: This study was approved by the ethical committee of Palacky University and complies with the current law of the Czech Republic.

Data accessibility: As a part of our policy all data associated with this paper will be publicly available on Dryad Digital Repository.

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