

# Hatching order explains an extrapair chick advantage in western bluebirds

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Within-family comparisons of offspring growth rates have provided important tests of genetic benefits of extrapair mating for females. Here, we demonstrate that hatching order explains the growth advantage for extrapair young in western bluebirds (*Sialia mexicana*); extrapair nestlings are larger than within-pair nestlings in the same nest, but they also hatch earlier in the clutch, thus benefiting from hatching asynchrony. By controlling for hatch order and other nongenetic factors and comparing mixed-paternity broods with genetically monogamous broods, we show that the extrapair nestling growth advantage is not genetically based. We cannot rule out the possibility that females benefit from extrapair mating because genetic quality indicators may appear later in life and may be independent of hatch order, however, based on our results, we do not see evidence of genetic superiority of extrapair nestlings. Although findings similar to ours have been attributed to maternal effects, it is currently unclear whether overrepresentation of extrapair nestlings early in the laying and hatching sequence results from investment patterns of females, their social mates, or extrapair males. This study highlights the need to investigate the potentially complex interactions among all players, and how these may lead to higher performance of extrapair offspring compared with within-pair offspring within the same family. *Key words*: extrapair paternity, genetic benefits, hatching asynchrony, hatching order, laying order, maternal effects. [*Behav Ecol* 21:802–807 (2010)]

The advent of molecular tools for genetic fingerprinting has made it apparent that males and females in socially monogamous species often mate outside of their pair bonds (e.g., in birds, Griffith et al. 2002; mammals, Cohas and Allaine 2009; reptiles, Uller and Olsson 2008; fish, reviewed by Avise et al. 2002). Several hypotheses have been proposed to explain how females benefit from having multiple mates, with the leading hypothesis being that females gain genetic benefits from mating with superior extrapair sires (Jennions and Petrie 2000). The genetic benefits hypothesis predicts that extrapair young should outperform young sired by the social mate (within-pair young) in such traits as growth, immune function, survival, and future reproductive success. Comparisons of the performance of extrapair and within-pair young raised in the same family control for such things as the environment, mother, and social father and assume that these factors account for most sources of nongenetic variation (Griffith et al. 2002). Early growth and survival of offspring are often used as proxies for future success because of the short-term nature of most studies and the difficulty of following offspring once they reach independence; differences in quality should also be pronounced at this point (reviewed in Akçay and Roughgarden 2007, but see Schmoll et al. 2003; Cohas et al. 2007). Evidence that extrapair young outperform their maternal half-siblings has therefore been interpreted as support for the idea that females select extrapair mates with superior or complemen-

tary genes (Kempnaers et al. 1997; Griffith et al. 2002; Schwagmeyer and Mock 2008).

Recent studies show that performance differences between extrapair and within-pair offspring are sometimes due to nongenetic factors that correlate with paternity. In 2 avian species, extrapair nestlings grew more quickly than nest mates sired by the social mate because they emerged earlier in the hatching sequence (blue tits, *Cyanistes caeruleus*, Magrath et al. 2009 and house wrens, *Troglodytes aedon*, Johnson et al. 2009). When extrapair nestlings are concentrated early in the brood sequence, their enhanced performance likely results not from genetic superiority but from the effects of hatching asynchrony favoring early-hatched young over later-hatched young (Magrath 1990; Duckworth 2009; Magrath et al. 2009). However, in 2 earlier studies, extrapair young tended to outperform within-pair young even when hatching order was controlled for (condition in collared flycatchers, *Ficedula albicollis*, Krist et al. 2004 and feather growth in tree swallows, *Tachycineta bicolor*, O'Brien and Dawson 2007). Only a few other studies have examined the relationship between paternity and hatching order, finding either no relationship (red-winged blackbirds, *Agelaius phoeniceus*, Westneat et al. 1995; tree swallows, *T. bicolor*; Barber and Robertson 2007 and O'Brien and Dawson 2007; and collared flycatchers, *F. albicollis*, Wilk et al. 2008) or that extrapair nestlings predominate early in the laying or hatching sequence (house sparrow, *Passer domesticus*, Cordero et al. 1999 and collared flycatchers, *F. albicollis*, Michl et al. 2002 and Krist et al. 2005). Clearly, additional studies are needed both to determine how frequently laying and hatching order are related to paternity and also to explore fully the possibility that effects of hatching order constitute evidence for female genetic benefits of extrapair mating.

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Received 26 October 2009; revised 15 March 2010; accepted 31 March 2010.

Besides hatching order and asynchrony, numerous other nongenetic factors can influence the relative growth of maternal half-siblings in birds. For example, one or both parents could preferentially feed offspring based on paternity. Egg size may also influence offspring growth if young from larger eggs become larger and have higher survival than those from smaller eggs (Amat et al. 2001; Whittingham et al. 2007). Females could vary the quality or quantity of egg contents, such as hormones and nutrients, across the laying sequence, conferring fitness advantages to extrapair offspring (Hargitai et al. 2006; Gil 2008). In birds, males are commonly larger than females; if extrapair young tend to be males, this would lead to a size difference between extrapair and within-pair nestlings (Trivers and Willard 1973; Janota et al. 2002).

By simultaneously considering egg volume, offspring sex, laying and hatching order, and paternity, we examined whether differences in nestling performance in western bluebirds (*Sialia mexicana*) stemmed from nongenetic factors as opposed to genetic differences between extrapair and within-pair nestlings. Unique to this study, we also examined the relative influence of nongenetic factors on nestling performance in monogamous broods to determine if all nestlings responded equally to their relative hatching order. Specifically, we predicted that if extrapair offspring are genetically predisposed to grow faster than the average offspring, then early hatching extrapair nestlings should be larger than early-hatching within-pair nestlings. On the other hand, if genetic effects are not important, then all nestlings should respond equally to nongenetic hatching order effects.

In this study, we demonstrate an extrapair nestling growth advantage and provide evidence that that this advantage stems from overrepresentation of extrapair young early in the hatching sequence. The early growth advantage we observed for extrapair young was also not distinguishable from that for early-hatching young in single-paternity broods. If overrepresentation of extrapair nestlings early in the brood is common, the next step is to distinguish who is driving the pattern (Kempnaers 2009). Although emphasis has been placed on maternal effects (Johnson et al. 2009; Magrath et al. 2009), we clarify the importance of considering alternative hypotheses that include the possibility that behaviors of the female's social mate, extrapair males, or the female herself drive the pattern (Westneat and Stewart 2003). We consider how investment by each player can influence the early positioning of extrapair nestlings and demonstrate how behavioral data may be used to distinguish among alternative investment patterns in this and future studies.

## MATERIALS AND METHODS

### Study system

Details of the long-term study of western bluebirds at Hastings Natural History Reserve, Carmel Valley, CA, can be found elsewhere (Dickinson et al. 1996), but briefly, in 2000 and 2001, we banded and identified breeders, monitored 89 nests for egg laying and hatching, and bled and banded 368 nestlings. We collected data only from a pair's first successful breeding attempt. Eggs were marked and measured as they were laid, and then beginning 1 day prior to estimated minimum date of hatch (average incubation period is 14 days but checks began on Day 11), we checked every hour to determine hatch order. Complete laying and hatching order were obtained for 30 broods and only partial hatch order for an additional 30 broods. Laying and hatching orders were transformed to a scale from 0 to 1 by dividing the actual order by the clutch or brood size. We then combined the transformed values into 5 relative laying or hatching sequence groups: 0.20, 0.40, 0.60,

0.80, and 1.0 based on the mean clutch size of  $4.78 \pm 0.06$  standard error (SE). Clutches and broods of 4 were binned into the groups 0.20, 0.40, 0.80, and 1.0. We calculated hatching asynchrony as the number of hours passing between hatching of the first and last nestling. Egg volume was calculated as  $([0.6057 - 0.0018 \times \text{width}] \text{length} \times \text{width}^2)$  (Narushin 2005). Within individuals, egg mass and volume were highly correlated (Pearson  $r = 0.92$ ,  $P < 0.001$ ,  $n = 307$ ). We weighed nestlings on the day they hatched and measured wing chord to the nearest 1 mm and mass to the nearest 0.5 g at 6 and 14 days after hatching. Nestling size at 14 days of age is a significant predictor of adult survival for male offspring (Dickinson 2004). Blood samples for paternity analysis were collected from nestlings on Day 6, and they were sexed using plumage on about Day 15.

### Microsatellite genotyping and parentage analysis

We extracted DNA from blood samples stored in lysis buffer using QiagenDNeasy tissue kits and then amplified fragments from 11 microsatellite regions with polymerase chain reaction (PCR) (*Smex* 2, 4, 5, and 7–14; Ferree et al. 2008). PCR products were sized on an Applied Biosystems 3100 DNA analyzer, and alleles were checked manually using GeneMapper software 3.7 (Applied Biosystems, Foster City, CA). In CERVUS 3.0, we calculated allele frequencies using the genotypes of all individuals in a given year and in the parentage analyses included all sampled males present in a given year as candidate fathers. The microsatellites had a mean of 12.82 alleles per locus and a combined probability of  $2.0 \times 10^{-5}$  of falsely assigning a putative father to an offspring given the mother's identity. Allele frequencies deviated from Hardy–Weinberg equilibrium at one locus, *Smex* 9, but the estimated frequency of null alleles was  $\leq 0.05$  for all loci. Based on detection of mismatches between offspring and known parents (mothers), we specified a typing error of 1%, likely due to mutation or low rates of null alleles at various loci. We specified the proportion of sampled candidate parents at 95% and based on known pedigrees included a relatedness of 0.375 among 1% of candidate parents. We accepted a putative male as the genetic sire for 332 of 339 genotyped offspring (98%) based on the following criteria: 1) a male was assigned at the 95% confidence interval for the comparison of the mother, father, and offspring genotypes (294 of 339 offspring); 2) no mother was genotyped, but the male was assigned at the 95% confidence interval for comparison of the father–offspring genotypes (36 of 339 offspring); or 3) no mother was genotyped, but the within-pair male was assigned at the 80% confidence interval for comparison of the father–offspring genotypes (2 of 339 offspring). Seven offspring were not assigned to any male and were categorized as extrapair. Finally, several families were excluded from the analyses because we both lacked a genotype for the mother and could not assign the offspring to particular sires.

### Statistical analyses

We calculated a mean mass and wing length separately for the extrapair and within-pair nestlings within each mixed-paternity brood and compared the means at 6 and 14 days of age using paired *t*-tests. To control for Type I error, multiple paired *t*-tests were treated by replacing alpha with  $d_i^* = (i/k) f_F$  for *i* of *k* comparisons to yield a false discovery rate,  $f_F = 0.05$  within the group of tests (Curran-Everett 2000). To determine how extrapair nestlings were distributed across the laying and hatching sequence, we used a generalized linear mixed model with a binomial error distribution and logit link, with paternity (1 = extrapair and 0 = within pair) as the response

variable and relative laying or hatching order and relative first egg date (centered around the median first egg date for a given year) as continuous fixed effects and family as a random effect. We tested for a sex bias in relation to paternity with a generalized linear model with binomial error distribution and logit link, with paternity (1 = extrapair and 0 = within-pair) as the response variable and sex (male or female) as a fixed effect.

We used general linear mixed models to test for differences in relative chick size in relation to sex (male or female), paternity (extrapair or within pair), and relative laying or hatching order as fixed effects, egg volume as a covariate, family as a random effect, and the interactions of sex with order and paternity. Because we were interested in within-brood variation in size, we calculated an individual's relative egg and chick size as the individual's value minus the nest's mean egg or chick size. This centering process essentially eliminated among-family variation (the effect of family identity as a random effect was zero in all of these models).

Finally, we were interested in comparing the growth rate of offspring from mixed-paternity broods with those from single-paternity broods to determine if hatching order had an equal effect on offspring size across broods. We used a general linear mixed model to determine if the relationship between laying or hatching position and relative offspring size differed for families with mixed- versus single-paternity by examining the interaction of paternity and order. Relative mass and wing length were the response variables, relative laying or hatching order whether broods were of mixed paternity and the interaction between sequence and brood paternity were fixed predictors, and family was included as a random effect.

In all tests, we removed nonsignificant interactions but retained the main effects in the final model. An individual's laying and hatch orders were significantly correlated ( $r = 0.74$ ,  $n = 201$ ,  $P < 0.001$ ), but we analyzed both to distinguish effects that stem from laying order (such as maternal investment in egg nutrients) from those associated with hatch order (such as the influence of hatch asynchrony). Means are presented with standard error of the mean unless otherwise noted. All tests were two-tailed and conducted with SPSS v16.0 or R v2.9.2.

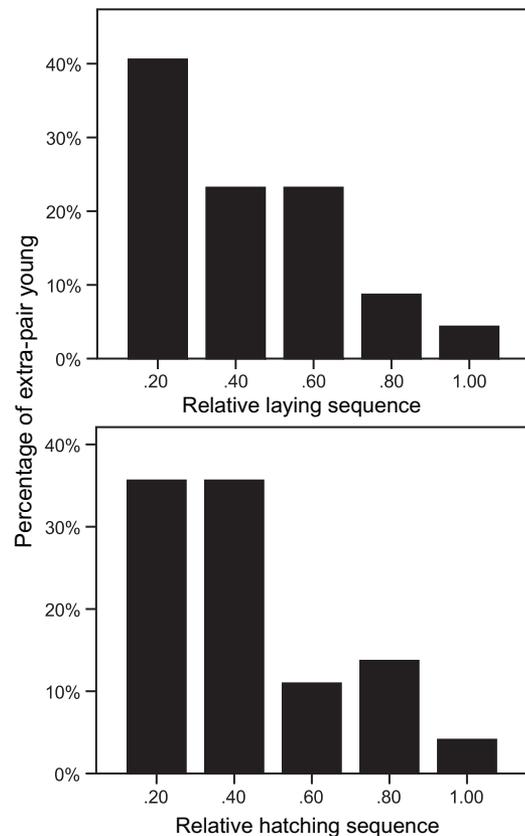
### Behavioral observations

Detailed observations of extrapair mating interactions are published elsewhere and were used to examine proximate mechanisms leading to the positioning of extrapair young early in the laying sequence (Dickinson and Leonard 1996; Dickinson 1997, 2001; Dickinson et al. 2000). Observations were made both under natural and experimental conditions, the latter involving 1-h detentions of resident males so that extrapair mating behavior could be observed without the chaos that normally ensues when the female's mate is present (Dickinson and Leonard 1996; Dickinson 1997; Dickinson et al. 2000). We divided the observations into 4 periods that reflect female fertility more than 10 days before egg laying began, 10 or fewer days prior to egg laying, during egg laying, and once incubation began. We assume that the first egg would be fertilized within the 10 days leading up to egg laying.

## RESULTS

### Comparison of extrapair and within-pair nestlings

Parentage analysis revealed extrapair nestlings in 39% of 89 broods, with 17.5% of 325 nestlings sired by extrapair males. Within broods of mixed paternity, the average extrapair nestling was significantly larger and heavier than the average



**Figure 1**

Percentage of 41 extrapair offspring distributed in relation to laying and hatching sequence in mixed-paternity broods. Laying and hatching orders were transformed to a scale from 0 to 1 by dividing the actual order by the clutch or brood size. We then combined the transformed values into 5 relative laying or hatching sequence groups: 0.20, 0.40, 0.60, 0.80, and 1.00 based on the mean clutch size of  $4.78 \pm 0.06$  SE (see "METHODS").

within-pair nestling at 6 days of age (Day 6 wing length, mean paired difference (mpd) =  $0.82 \pm 0.33$  mm, paired  $t = 2.46$ , degrees of freedom [df] = 33,  $P = 0.02$ ,  $d_i^* = 0.025$  and mass, mpd =  $0.74 \pm 0.32$  g, paired  $t = 2.33$ , df = 33,  $P = 0.03$ ,  $d_i^* = 0.035$ ) and significantly larger but not significantly heavier at 14 days of age (Day 14 wing length, mpd =  $2.32 \pm 0.56$  mm, paired  $t = 4.40$ , df = 29,  $P < 0.001$ ,  $d_i^* = 0.013$  and mass, mpd =  $0.59 \pm 0.32$  g, paired  $t = 1.81$ , df = 29,  $P = 0.08$ ,  $d_i^* = 0.05$ ).

We then examined several nongenetic factors that could influence nestling size, specifically egg volume, nestling sex, laying order, and hatching order. Despite becoming larger nestlings, extrapair nestlings tended to come from smaller eggs than within-pair nestlings (egg volume, mpd =  $-53.6 \pm 31.02$  mm<sup>3</sup>, paired  $t = -1.73$ , df = 25,  $P = 0.096$ ). Extrapair nestlings were not more likely to be male than female, as would be predicted if sexual dimorphism explained the nestling size difference or if maternal effects conferred growth advantages to early sons (Wald  $\chi^2 = 1.47$ , df = 1,  $P = 0.23$ ). In relation to order placement, however, we found that extrapair nestlings were significantly overrepresented early in the laying and hatching sequence (laying sequence:  $z = -3.46$ ,  $P < 0.001$  and hatching sequence:  $z = -3.51$ ,  $P < 0.001$ ; Figure 1).

In a single model, we then investigated the relative contributions of nestling sex, paternity, laying and hatching order, and relative egg volume to the relative size of nestlings. At 6 and 14 days of age, nestlings found earlier in the laying and hatching sequence weighed more and had longer relative wing lengths

**Table 1**  
Effects on relative nestling size at 6 and 14 days of age

	Day 6 mass	Day 6 wing length	Day 14 mass	Day 14 wing length
Laying order				
Sex	0.25 (1, 83)	1.21 (1, 83)	3.48 (1, 83) <sup>+</sup>	0.30 (1, 83)
Paternity	2.06 (1, 83)	3.86 (1, 83) <sup>+</sup>	0.55 (1, 83)	1.87 (1, 83)
Laying order	9.93 (4, 83) <sup>***</sup>	17.48 (4, 83) <sup>***</sup>	3.03 (4, 83) <sup>*</sup>	9.48 (4, 83) <sup>***</sup>
Egg volume	0.62 (1, 83)	0.21 (1, 83)	0.002 (1, 83)	0.31 (1, 83)
Hatching order				
Sex	0.005 (1, 86)	0.74 (1, 86)	2.28 (1, 86)	0.92 (1, 86)
Paternity	1.72 (1, 86)	3.79 (1, 86) <sup>+</sup>	0.57 (1, 86)	1.93 (1, 86)
Hatching order	13.026 (4, 86) <sup>***</sup>	17.45 (4, 86) <sup>***</sup>	1.64 (4, 86)	12.60 (4, 86) <sup>***</sup>
Egg volume	0.45 (1, 86)	0.003 (1, 86)	0.001 (1, 86)	0.31 (1, 86)

Displayed are *F* values (with *df* in parentheses) from general linear mixed models testing the effect of sex (male and female), paternity (within-pair and extrapair), relative laying or hatching order, and relative egg volume on day 6 and day 14 mass and wing lengths. Symbols indicate level of significance as follows: <sup>+</sup>*P* < 0.10, <sup>\*</sup>*P* < 0.05, <sup>\*\*\*</sup>*P* < 0.001.

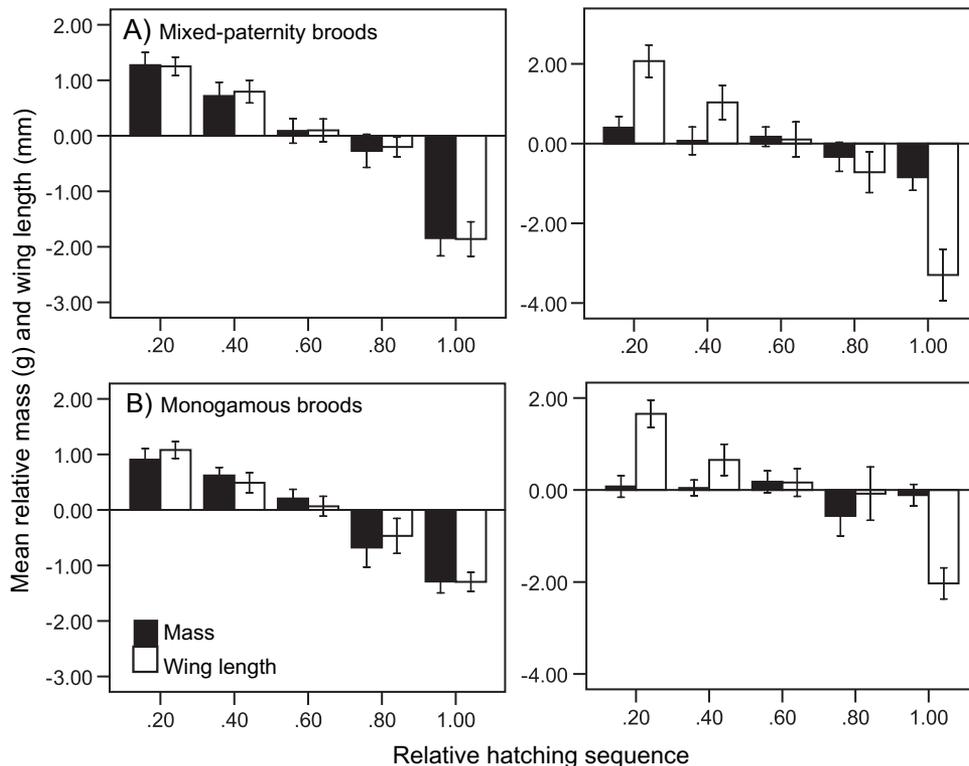
than those in later positions, whereas paternity, sex, and relative egg volume did not significantly affect nestling size (Table 1; Figure 2a). These analyses demonstrate that laying and hatching order within the nest largely accounts for superior performance, measured as relative nestling size, of extrapair nestlings and therefore do not support the genetic benefit hypothesis.

**The role of order**

To further examine the role of hatching order in determining nestling size, we compared the relationship between order and offspring size in mixed- versus single-paternity broods. We found that the effects of laying and hatching order on relative offspring size within a brood were not significantly different between broods with and without extrapair paternity, specifically the interaction term of paternity and order was not significant (Table 2; Figure 2a,b). This result demonstrates that

position in the hatching order has the same influence on nestling size regardless of paternity status of the individual or the brood.

If hatching sequence largely accounts for the size difference between extrapair and within-pair nestlings, the magnitude of the size difference should depend on the extent of hatching asynchrony. The mean hatching interval between first and last eggs was 19.14 h ( $\pm 1.13$ , *n* = 50) and was nearly identical for broods with and without extrapair paternity ( $19.34 \pm 1.90$  h vs.  $19.11 \pm 1.54$  h, *F*<sub>1, 44</sub> = 0.01, *P* = 0.93). As expected, the magnitude of the difference in mass and wing length between first and last nestlings increased with degree of hatching asynchrony when measured at 6 and 14 days of age (Day 6 mass, *r* = 0.56, *n* = 26, *P* = 0.003; wing length, *r* = 0.51, *n* = 26, *P* = 0.007 and Day 14 mass, *r* = 0.38, *n* = 25, *P* = 0.06; wing length, *r* = 0.54, *n* = 25, *P* = 0.006). The size advantage conferred from hatching asynchrony did not stem from initial maternal investment favoring early eggs or young because the relationship



**Figure 2**  
Mean relative mass and wing length versus relative hatching sequence for A) 108 young from mixed-paternity and B) 137 young from monogamous broods at 6 days (left panels) and 14 days of age (right panels). Bars represent mean (+1SE) of relative mass (black bars) and wing length (open bars). Relative values were calculated as an individual's mass or wing length minus the mean value in that individual's broods.

**Table 2**  
**Comparison of relationship between offspring size and laying or hatching order in mixed- versus single-paternity broods**

	Day 6 mass	Day 6 wing length	Day 14 mass	Day 14 wing length
Laying order				
Mixed/single paternity	0.19 (1, 184)	0.31 (1, 184)	0.03 (1, 178)	0.10 (1, 176)
Laying order	32.10 (4, 184)***	43.44 (4, 184)***	4.79 (4, 178)***	26.98 (4, 176)***
Interaction	0.93 (4, 184)	1.56 (4, 184)	1.55 (4, 178)	0.83 (4, 176)
Hatching order				
Mixed/single paternity	0.06 (1, 190)	0.09 (1, 190)	0.001 (1, 184)	0.44 (1, 182)
Hatching order	43.63 (4, 190)***	59.57 (4, 190)***	3.22 (4, 184)*	36.20 (4, 182)***
Interaction	1.41 (4, 190)	1.61 (4, 190)	1.51 (4, 184)	1.72 (4, 182)

Displayed are *F* values (with df in parentheses) from general linear mixed models testing the effect of paternity (mixed- versus single-paternity), relative laying or hatching order, and an interaction of paternity and order on day 6 and day 14 mass and wing lengths. Symbols indicate level of significance as follows: \**P* < 0.05, \*\*\**P* < 0.001.

between size and asynchrony did not exist when young were weighed on the day of hatching (*P* > 0.20). In terms of laying sequence, an additional advantage of early egg placement was the high hatch rate of first eggs (100%): in the 10 (of 89) nests where 1 egg failed to hatch, it was always a penultimate or last egg.

## DISCUSSION

As predicted by the genetic benefits hypothesis, extrapair nestlings were larger than their half siblings at 6 and 14 days of age, but this difference was best explained by the overrepresentation of extrapair young early in the laying and hatching sequence. Other nongenetic factors that could influence nestling size did not appear to account for our results: Extrapair nestlings were not male biased and tended to come from relatively small eggs. Furthermore, preferential feeding of extrapair young by females probably did not explain the size difference because parents do not tend to discriminate among young based on parentage, even in cross-fostering experiments (Leonard et al. 1995 for this population; Westneat et al. 1995; Kempenaers and Sheldon 1996). We also found that hatching order had similar effects on offspring size in both mixed-paternity and monogamous broods. Hatching asynchrony appears to be responsible for the primary benefit of early positioning, by giving early-hatched young a head start in feeding and growth. From these data, we conclude that within-pair–extrapair comparisons of nestling growth do not necessarily reflect genetic benefits of extrapair mating if nongenetic influences on fitness, particularly hatching order, are not controlled for (Johnson et al. 2009; Magrath et al. 2009). Lack of support for genetic benefits within the nestling stage does not refute the possibility of genetic benefits expressed later in life; however, the effects that hatching order has on nestling growth likely affect long-term survival (Kissner and Weatherhead 2005; Medeiros and Freed 2009). Future studies should investigate the relative influences of hatching order and paternity on lifetime fitness of extrapair and within-pair offspring.

### Looking beyond maternal effects

Although maternal effects are one potential explanation for early placement of extrapair young (Johnson et al. 2009; Magrath et al. 2009), the benefits of such a pattern for female western bluebirds are not yet clear. If females gain from early placement of extrapair nestlings within the clutch sequence, we would also expect them to bias their investment in early eggs or via sex allocation, patterns we did not find. Based on our data, genetic differences in quality between extrapair and

within-pair young are still possible; however, they are not apparent in the nestling stage. For females to benefit from early placement, extrapair young would have to exhibit enhanced fitness later in life, and this fitness would have to be tied to genetic differences rather than simply to hatch order. For this reason, additional data are required to address the fitness consequences for females of early placement of extrapair young. Furthermore, given that paternity is the result of interactions among the female, the extrapair male, and her mate (Westneat and Stewart 2003), we cannot take for granted the possibility that females are driving the pattern.

For example, it is possible that extrapair males are driving the placement of their young early in the brood to gain phenotypic advantages that hatching asynchrony confers to early eggs. This hypothesis is consistent with the observation that in western bluebirds, extrapair males most commonly intrude on neighboring territories during the period 10 days prior to the target female's onset of laying (Dickinson and Leonard 1996). This behavior alone, however, is not likely to account for the early positioning of extrapair young in broods because extrapair copulations are far rarer than within-pair copulations, even during the prelaying period (Dickinson and Leonard 1996; Dickinson 1997; Dickinson et al. 2000). In other words, the rates of extrapair paternity and the position of extrapair young early in the clutch sequence cannot be explained by the extrapair male's copulation rate just prior to egg laying. The extrapair males' effort would have to be augmented through increased success in sperm competition or preferential use of extrapair sperm by females (Pizzari et al. 2003).

Finally, the role of the within-pair male in positioning extrapair young early in the clutch sequence is not clear. Within-pair males begin to attempt to copulate with females as much as 60 days prior to laying and do not appear to diminish their attempt rate until the day after the last egg is laid (Dickinson and Leonard 1996). Females begin accepting about 80% of within-pair copulations in the 10 days prior to laying and continue this high rate of acceptance during egg laying (Dickinson and Leonard 1996). Mate guarding by within-pair male's can also influence paternity (Chuang-Dobbs et al. 2001). Although within-pair males vary their mate guarding to match the female's receptive period, they do not appear to guard or copulate with their mates less before than during laying (Dickinson and Leonard 1996). We therefore see no behavioral evidence on the part of within-pair males that would reduce their paternity in early-laid eggs.

In all, these results suggest that the early placement of extrapair young likely stems from the temporal pattern of investment in extrapair copulations by extrapair males in conjunction with some unknown mechanism that make them

disproportionately successful at sperm competition either through increased competitive ability of extrapair over within-pair males' sperm or through increased acceptance and use of extrapair males' sperm by females. Teasing apart male and female roles is challenging; however, future extension of our data to examine long-term survival and reproductive success of extrapair and within-pair nestlings will help to elucidate whether there are fitness advantages of early placement of extrapair young for females or whether, instead, those advantages are conferred on extrapair males alone.

## FUNDING

National Science Foundation-Integrative Organismal Systems (0097027 and 0718416 to J.D.); the Natural Sciences and Engineering Research Council of Canada (W.R.).

We thank W. Hochachka and B. Zuckerberg for statistical advice; R. Naka and C. Makarewich for lab assistance; and A. Chaine, K. Holder, B. Kempnaers, W. Koenig, R. Lohnes, B. Lyon, T. Phillips, G. Rosenthal, and D. Shizuka for helpful comments. We also thank staff of the Museum of Vertebrate Zoology and Hastings Reserve for logistical support and landowners at Oak Ridge Ranch for access to their property. Many field interns helped keep the population banded over the years, leading to this study.

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