

SHORT COMMUNICATION

Within-clutch variation in offspring sex determined by differences in sire body size: cryptic mate choice in the wild

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Abstract

Sexual selection theory predicts that paternal quality should drive female investment in progeny. We tested whether polyandrous female side-blotched lizards, *Uta stansburiana*, would adjust within-clutch progeny investment according to sire phenotypes. In two different years, polyandrous females selectively used sperm from larger sires to produce sons and used sperm from smaller sires to produce daughters. This cryptic sperm choice had significant effects on progeny survival to maturity that were consistent with sexually antagonistic effects associated with sire body size. Large sires produced sons with high viability and small sires produced daughters with high viability. These results are consistent with our previous findings that alleles for male body size have different fitness effects in male and female progeny. Breeding experiments in the laboratory indicate that results from the wild are more likely due to female choice than biased sperm production by males. Our results demonstrate highly refined gender-specific female choice for sperm and indicate that sire body size may signal the quality of sons or daughters that a sire will produce.

Introduction

Cryptic female choice (Eberhard, 1996; Jones, 2002) is one of the most controversial means by which females select mates. Cryptic choice may reflect differential sperm choice by females and/or sperm competition among males, a distinction that has received considerable debate (Birkhead, 1998; Eberhard, 2000; Kempnaers *et al.*, 2000). Growing evidence indicates that in cases where females mate with multiple males (polyandry), cryptic choice of different males' sperm can have important fitness consequences for sires, dams and their progeny (Gowaty, 1997; Jennions & Petrie, 2000). Polyandrous females have an increased opportunity to engage in cryptic mate choice and theory suggests that the potential genetic benefits associated with cryptic choice may outweigh the costs of female promiscuity (Yasui, 1997).

Empirical studies with reptiles have demonstrated that cryptic sperm choice benefits polyandrous females through inbreeding avoidance (Madsen *et al.*, 1992; Olsson *et al.*, 1994), which increases offspring viability. Females can also benefit by biasing sex ratios as a function of mate quality, and evidence for sex ratio adjustment has been demonstrated in a surprising number of studies (e.g. Svensson & Nilsson, 1996; Sheldon *et al.*, 1999; West & Sheldon, 2002). In most of these studies, females overproduce one or the other sex depending on environmental conditions (Komdeur *et al.*, 1997; Radford & Blakey, 2000) or sire quality (West & Sheldon, 2002) [including survival prospects (Svensson & Nilsson, 1996), sexual ornaments (Ellegren *et al.*, 1996) age (Mange, 1970) and male body size (Kolliker *et al.*, 1999)]. In contrast, here we demonstrate a new form of cryptic choice for good genes (Petrie, 1994) in which polyandrous females differentially allocate X and Y sperm from small and large males, respectively, to produce both highly viable sons and daughters within a clutch.

Female side-blotched lizards are among the most promiscuous amniote vertebrates studied; up to 81% of

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all clutches are fertilized by multiple males (Zamudio & Sinervo, 2000). Lizard population densities cycle with a 2-year periodicity that favours large female progeny at high density and small female progeny at low density (Sinervo *et al.*, 2000). Side-blotched lizards also exhibit extreme sexual size dimorphism in which males are 60% larger than females. Males are characterized by one of three genetically determined throat colours (orange, blue, yellow) each associated with alternative mating and territorial behaviours that are considered in detail elsewhere (Sinervo & Lively, 1996; Calsbeek *et al.*, 2002). In this study, we focus on the contribution of male body size to the relative fitness of sons and daughters irrespective of morphotype.

Male body size has important consequences for territoriality and reproductive success (Calsbeek *et al.*, 2002), and is highly heritable (data from this study: $h^2 = 0.73 \pm 0.26$, $F_{1,18} = 7.559$, $P < 0.01$). Moreover, we have shown elsewhere that genes for male body size are genetically correlated with life-history traits in females (e.g. laying date and egg mass; Sinervo and Calsbeek, submitted). These genetic correlations lead to alternative fitness optima in the two sexes (Chippindale *et al.*, 2001). For example, in another study (Sinervo and Calsbeek, submitted) natural selection on male body size alleles was weakly stabilizing in sons (generally favouring larger males), whereas the female traits genetically correlated with male body size (i.e. laying date and egg mass) were under strong disruptive selection in daughters. Alternative fitness topologies in male and female progeny are the result of sexual conflict (Chippindale *et al.*, 2001; Rice & Chippindale, 2001). That is, although alleles that enhance male body size are a benefit in male-male competition, they could be harmful for females if the sexes have different body size optima. Because of the negative genetic correlations between sire body size alleles and life-history traits expressed in daughters, potential differences in the genetic quality of male and female progeny may be due to a sire's body size.

Consequently, our expectation in this study is that genes for sire body size should contribute differentially to the fitness of sons and daughters. Sexual selection pulls body size alleles in one direction (e.g. favouring larger male body size), whereas antagonistic natural selection on female life-history traits pulls those same alleles in the opposite direction (e.g. favouring earlier laying dates). Together, these genetic correlations and divergent selection pressures subject females to the chronic forces of ontogenetic conflict.

One way for females to mitigate conflict is by sorting male benefit/female detriment genes into sons and female benefit/male detriment genes into daughters. This prediction has been supported by previous work (Calsbeek & Sinervo, 2002a), where female lizards chose to reside on high quality territories but mated with both small resident males and their larger male neighbours from low quality territories. Females that mated with

multiple males differentially allocated sperm from small males to make daughters and from large males to make sons (Calsbeek & Sinervo, 2002a).

In this study, we report paternity results from an additional independent year, and include data on progeny survival to maturity as a function of sire size. We also report results from a laboratory breeding study to show that our results are most consistent with the interpretation that females perform post-copulatory manipulations of sperm to produce high quality sons and daughters. We used the high incidence of multiple paternity in this lizard population to further test the prediction that females would alter sex allocation based on differences in sire body size. We estimated paternity (using DNA microsatellites) and survival to maturity for 75 first-clutch progeny from 20 polyandrous females during 1999 and 2001. In both years, our study populations were in the low-density phase of their 2-year population density cycle – a condition that favours small female progeny (Sinervo *et al.*, 2000). Paternity and survival data presented in our analyses are from a long-term experimental study where territory quality was manipulated on replicate sandstone outcroppings (Calsbeek *et al.*, 2002; Calsbeek & Sinervo, 2002b). Manipulations are not likely to have played a causative role in this study, as the range of territory quality on experimental plots is the same as in natural variation (Calsbeek & Sinervo, 2002b). Furthermore, rates of multiple paternity are 81% in natural variation and 78% on the experimental plots. Side-blotched lizards in our study population are an annual species (Sinervo *et al.*, 2000) and most individuals die after a single reproductive season. Thus, our study encompasses two unique cohorts of sires, dams and progeny.

Methods

Sex allocation

Analyses of sex ratio variation are fraught with controversy concerning appropriate statistical methods (Krackow & Tkadlec, 2001). Thus, it is imperative to stress here that our study does not pertain to overall brood sex ratios *per se*. The focus of the present study concerns the effects of relative sire body size on the production of sons and daughters within a clutch. However, we do not address issues relating to sex ratio adjustment. We measured differences in the production of sons and daughters within clutches from individual females using a multivariate ANOVA (Pilai-Trace) in which the dependent variables, numbers of sons and daughters, were tested against the independent factor 'sire body size'. All polyandrous females in our analyses laid clutches of eggs that were co-sired by two males. This allowed us to unambiguously categorize the relative body mass of the two sires as factors ('large' vs. 'small'). We calculated the relationship between offspring gender and sire body size

(nested within females) testing the specific prediction that larger sires would produce sons and smaller sires would produce daughters (Calsbeek & Sinervo, 2002a). This prediction was based on our previous observation that alleles for male body size have sexually antagonistic effects in sons and daughters and that natural and sexual selection should favour a female's ability to sort these alleles into progeny of the appropriate gender (Sinervo and Calsbeek, submitted).

Estimating survival to maturity

Maternity was known with certainty because we captured all 20 gravid dams from the population to lay eggs in the laboratory (mean clutch size = 4.5 eggs). Females were released to their spot of capture after laying eggs, and eggs were incubated under standard conditions (Sinervo *et al.*, 2000). Progeny were sexed at hatching [chromosomal sex determination (Reeder & Wiens, 1996), males have enlarged post-anal scales] and released randomly with respect to their mother's territory within 3 days of hatching. We released progeny in pairs (determined randomly with respect to sibship) to sites from which we had captured gravid females earlier during the summer. As a means of reducing the potential influence of maternal effects, no progeny were released to their maternal nest site. We measured hatching to adult survivorship by recapturing all juveniles the following year during spring and through the end of summer (the life-span of most side-blotched lizards). In addition to the focal study site where progeny were released, we also monitored all surrounding lizard habitat out to a distance of at least 1.5 km to record dispersal events (Sinervo *et al.*, 2000). These adjacent areas are studied in other experiments and are thus monitored with the same intensity as the focal study site in this experiment. All lizards not captured during our censuses were considered to have died. Survival data are distributed binomially (live/die) and we tested for the effects of sire body size on progeny survival using logistic regression.

Heritability

Estimates for the heritability of male body size were calculated from $n = 20$ sire/son pairs collected from a separate outcropping in the wild. The h^2 estimate 0.73 ± 0.26 is based on the residuals of 4 years of body size data removing any effects caused by variation in year. However, even without removing year effects, we still measured a significant heritability for male body size ($h^2 = 0.62$, $F_{1,18} = 4.819$, $P < 0.04$).

DNA paternity

We extracted genomic DNA from toe tissue by overnight incubation at 55 °C in 500 μ L 5% Chelex (Bio-Rad,

Hercules, CA, USA) and 2 μ L proteinase K solution (20 mg mL⁻¹). The extract was centrifuged and diluted 1 : 10. We amplified nine microsatellite loci from this extract via polymerase chain reaction. We assessed length polymorphism with fluorescently labelled forward primers on an automated DNA sequencer (ABI 3100, Applied Biosystems, Foster City, CA, USA). We assigned paternity using the program Kinship (Goodnight *et al.*, 1996) that uses maximum likelihood and incorporates exclusionary criteria. Paternity results using Kinship and other software are similar (Zamudio & Sinervo, 2000), and we report only the results from Kinship. We accepted a male as a hatchling's sire if the likelihood of paternity was significantly different from that expected for unrelated males ($P < 0.05$). We used detailed territory maps to further enhance our ability to estimate paternity. We subdivided the site into neighbourhoods separated by unsuitable adult habitat and searched for putative sires within the female's neighbourhood. We assigned as sire the male with highest significant likelihood value in this neighbourhood. We also screened male genotypes in a female's adjacent and immediate neighbourhoods to account for males that may have moved between neighbourhoods (an extremely rare occurrence; Zamudio & Sinervo, 2000). Males from this larger area were assigned as sire only if their likelihood values were higher than those for males in the female's immediate neighbourhood.

Sire effects on sperm production

We tested whether males manipulated sperm production as a function of their body size in two different ways. First, we measured the repeatability of progeny sex as a function of sire body size using males that reproduced with multiple females. Repeatability (or intra-class correlation) measures the correlation between sex ratios produced by the same male with two different females (Lessells & Boag, 1987). We scored the gender of progeny sired by 10 sires from the field data, each of which sired progeny with two different females (Kempnaers *et al.*, 2000; Clark, 2002). High (significant) repeatability would indicate that individual sires produce similar sex ratios with all females (e.g. sires produce sons or daughters in all reproductive episodes). Low repeatability would suggest that sires do not always produce the same ratio of sons to daughters.

Secondly, we performed controlled matings in the laboratory (restricted to monogamy) by pairing individual males and virgin females in a terrarium during the female's receptive period. We can reliably score virgin females in the wild by palpating follicles early during the reproductive cycle (Sinervo *et al.*, 2000). Because *Uta* is an annual lizard, all females were entering their first reproductive episode and had not mated prior to our experiment. Matings were videotaped for behavioural analyses to be published elsewhere (C. Bleay, in preparation).

Results

Paternity analyses revealed that sire body size had significant effects on the production of sons and daughters within clutches of eggs produced by polyandrous females. Sperm from larger sires was significantly more likely to produce sons, and the sperm from smaller sires was more likely to produce daughters (multivariate ANOVA; sire effects nested within females, $F_{1,32} = 6.52$, $P < 0.05$; effect of year n.s.; Fig. 1). These effects were also significant in individual years (sex \times sire effect during 1999: $F_{1,12} = 7.68$, $P < 0.05$; sire effect during 2001: $F_{1,20} = 4.32$, $P = 0.05$; Fig. 1).



Fig. 1 The mean number of progeny (sons and daughters) sired by large vs. small males within a female's clutch during 1999 and 2001. In both years, the production of sons and daughters depended on relative sire body size. Females were more likely to produce sons with the sperm from relatively larger sires in both years. During 1999, females were also significantly more likely to produce daughters with the sperm from relatively smaller sires. Histogram bars represent the mean numbers of sons and daughters ± 1 SE.

Moreover, cryptic female choice of X and Y sperm from different males suggests that sperm sorting has important effects on progeny viability. Survival of sons and daughters differed significantly depending on sire body size. Sons had higher survival to maturity when sired by larger males and daughters had higher survival to maturity when sired by smaller males (interaction term from logistic regression: sire body size \times progeny sex, $r^2 = 0.19$, $P < 0.05$; Fig. 2). Progeny survival to maturity was not significantly affected by paternal genotype (throat score, n.s.), territory quality (n.s.), or maternal effects (e.g. egg size, n.s.; laying date, n.s.).

Our results indicate that either sires or dams can influence the sex of progeny produced within a clutch. One interpretation of our results is that sires adjusted the production of sons and daughters within clutches of eggs. For example, it is possible that sire body size affected sperm production. Testis size scales with male body size

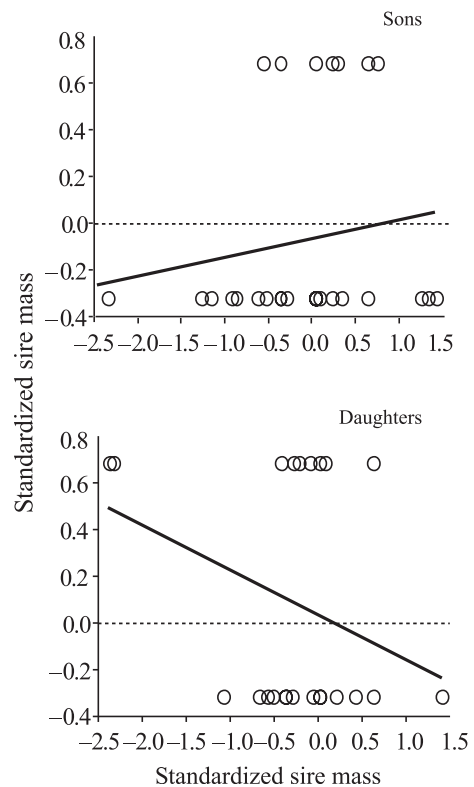


Fig. 2 Progeny sex allocation had significant effects on survival. During 1999 and 2001 significant differences in viability selection (survival to maturity) favoured sons that were sired by relatively larger males and daughters that were sired by relatively smaller males. Considering only progeny of polyandrous females, we recaptured four of 36 progeny during 1999 and 17 of 39 progeny during 2002 that had been released during the previous fall, and to whom we could accurately assign sires with paternity analyses. Sire body size and survival have been standardized to mean zero with unit variance (Arnold & Wade, 1984).

(Olsson *et al.*, 1996) and large males may have simply sired more offspring (irrespective of progeny gender) by producing more sperm. This was not the case (ANOVA: $F_{1,34} = 0.63$, n.s.). Alternatively, large males may have produced more Y sperm, or Y sperm that were more motile or more viable compared with smaller males. If males overproduce either X or Y sperm as a function of their body size, then a male that sires sons with one mate should sire sons with all of his mates. However, the repeatability of offspring sex as a function of sire body size was not significant ($F_{9,10} = 1.103$, repeatability = 0.046; n.s.), because males produce sons when relatively large and daughters when relatively small compared with other males that co-sire the females clutch.

The relative numbers of sons and daughters produced by 41 females bred in the laboratory with a single sire did not differ as a function of sire body size (multivariate ANOVA: sire body size nested within females, $F_{1,39} = 0.974$, n.s.). Thus, we find no evidence that X and Y sperm production is body size dependent, which might arise if body size were associated with a segregation distorter (Wilkinson & Reillo, 1994; Wilkinson *et al.*, 1998).

Discussion

We have demonstrated that the sex of progeny within a female's clutch depends on the relative body sizes of different sires. Although we cannot definitively rule out an effect of biased sperm production or fertilization success, our data from both the field and laboratory breeding experiments suggest that offspring production is not under male control. An alternative interpretation that is completely consistent with the survival data is that females differentially allocate sire genes to male and female progeny in a form of cryptic mate choice. Selective use of sperm from different sires to produce sons and daughters suggests that females sort sperm based on both sire identity and the sex chromosome (X or Y) carried by the sperm. The use of a male's sperm to produce sons when relatively large (relative to the co-siring male) but daughters when relatively small should not necessarily be interpreted to mean females are forced to allocate sperm maladaptively. Instead, we suggest that just as male body size varies continuously, so too does the degree of sexual conflict imposed by male body size alleles. This explains the nonrepeatability of offspring sex as a function of male body size. Females must always make sperm allocation decisions based on the relative size of sires, and this decision will be constrained by information from the female's local neighbourhood (typically two to three males; see Sinervo *et al.*, 2000). Although progeny quality may not always be optimal, progeny fitness will always be highest when females make reproductive decisions based on relative male body size.

Results from controlled monogamous matings in the laboratory indicate that males do not bias sperm production as a function of their body size. However, the lack of a relationship between sire size and the sex of progeny from these matings is still surprising, as we would have predicted that females would have been able to gauge sire size and hence project the quality of sons vs. daughters that he would produce. That this was not the case indicates either that females can only determine sire size in a relative context (e.g. compared with another male with which she has mated) or that some other process is driving the pattern of progeny gender allocation. In any case, based on the repeated measurement of the same effects in two separate years, and that results are in line with our predictions concerning sexual conflict, we conclude that the allocation of progeny gender is subject to selection.

The effects of sire body size on the viability of sons and daughters (Fig. 2) are also consistent with our previous results demonstrating ontogenetic conflict (Sinervo and Calsbeek, submitted). Sample sizes are relatively small in our selection analysis because we consider only the survival of progeny from clutches co-sired by two males. These data must therefore be interpreted cautiously. However, the difference in survival between sons and daughters is significant despite this limitation and is moreover, in line with the survival affects predicted under the conditions of sexual conflict. In another study (Sinervo and Calsbeek, submitted), directional sexual selection and stabilizing natural selection on male body size both favoured generally larger males, whereas natural selection on the genetically correlated female life-history traits, egg mass and laying dates, was disruptive. These alternative fitness topologies indicate that alleles for male body size have different effects on the fitness of male and female progeny. Large sire body size indicates that a male carries male benefit/female detriment alleles whereas the converse is true of small sires, which carry female benefit/male detriment alleles (Chippindale *et al.*, 2001). This difference reinforces the fundamental sexual dimorphism between male and female side-blotched lizards. Ultimately, stabilizing selection on male body size will place an upper limit on this dimorphism and prevent a runaway (Fisher, 1930).

One obvious shortcoming of this study is our inability to present the physiological mechanism whereby females sort sperm from different sires. Evidence for sperm choice of a different type has previously been demonstrated in these (Zamudio & Sinervo, 2000) and other lizards (Olsson *et al.*, 1996) as well as in crickets (Tregenza & Wedell, 2002). Female side-blotched lizards are known to store sperm for use in future reproductive episodes, and may do so for considerable time periods (e.g. 2 months; Zamudio & Sinervo, 2000). Whether females can also screen X and Y sperm haplotypes is still uncertain, but such a result is strongly implicated by our data. Age-dependent sorting of X and Y bearing sperm has been

reported in *Drosophila* (Mange, 1970) and precise sperm sorting (X vs. O) has also been recently reported in spiders (Aviles *et al.*, 2000). Finally, sperm recognition is facilitated in many species by just a few genes expressed as protein signals on the gamete surface (Vacquier, 1995).

When genes for male and female viability have alternative expression in sires (e.g. large vs. small adult body size) females will be under strong selection to sort sperm from different sires into male and female progeny. Indeed, female side-blotched lizards appear to sort sperm to produce high quality sons from large sires and high quality daughters from small sires. Our results suggest that selection has operated on female side-blotched lizards to mitigate ontogenetic conflict (Chippindale *et al.*, 2001) between body size alleles in sires and genetically correlated life-history traits in females (Sinervo and Calsbeek, submitted).

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References

- Arnold, S.J. & Wade, M.J. 1984. On the measurement of natural and sexual selection: theory. *Evolution* **38**: 709–719.
- Aviles, L., McCormack, J., Cutter, A. & Bukowski, T. 2000. Precise, highly female-biased sex ratios in a social spider. *Proc. R. Soc. Lond. B* **267**: 1445–1449.
- Birkhead, T.R. 1998. Cryptic female choice: criteria for establishing female sperm choice. *Evolution* **52**: 1212–1218.
- Calsbeek, R. & Sinervo, B. 2002a. Uncoupling direct and indirect benefits of mate choice in the wild. *Proc. Natl. Acad. Sci. USA* **99**: 14897–14902.
- Calsbeek, R. & Sinervo, B. 2002b. An experimental test of the ideal despotic distribution. *J. Anim. Ecol.* **71**: 513–523.
- Calsbeek, R., Alonzo, S.H., Zamudio, K. & Sinervo, B. 2002. Sexual selection and alternative strategies generate demographic stochasticity in small populations. *Proc. R. Soc. Lond. B* **269**: 157–164.
- Chippindale, A.K., Gibson, J.R. & Rice, W.R. 2001. Negative genetic correlation for adult fitness between sexes reveals ontogenetic conflict in *Drosophila*. *Proc. Natl. Acad. Sci. USA* **98**: 1671–1675.
- Clark, A.G. 2002. Sperm competition and the maintenance of polymorphism. *Heredity* **88**: 148–153.
- Eberhard, W.G. 1996. *Monographs in Behavior and Ecology: Female Control: Sexual Selection by Cryptic Female Choice*. Princeton University Press Technical Books, Princeton, NJ.
- Eberhard, W.G. 2000. Criteria for demonstrating postcopulatory female choice. *Evolution* **54**: 1047–1050.
- Ellegren, H., Gustafsson, L. & Sheldon, B.C. 1996. Sex ratio adjustment in relation to paternal attractiveness in a wild bird population. *Proc. Natl. Acad. Sci. USA* **93**: 11723–11728.
- Fisher, R. 1930. *The Genetical Theory of Natural Selection*. Oxford University Press, Oxford.
- Goodnight, K.F., Quellar, D.C. & Poznansky, T. 1996. Kinship v1.1.2. Goodnight Software (distributed by author).
- Gowaty, P.A. 1997. Sexual dialectics, sexual selection, and variation in mating behavior. In: *Feminism and Evolutionary Biology* (P. A. Gowaty, ed.), pp. 351–384. Chapman Hall, New York.
- Jennions, M.D. & Petrie, M. 2000. Why do females mate multiply? A review of the genetic benefits. *Biol. Rev. (Camb.)* **75**: 21–64.
- Jones, A. 2002. The evolution of alternative cryptic female choice strategies in age-structured populations. *Evolution* **56**: 2530–2536.
- Kempnaers, B., Foerster, K., Questiau, S., Robertson, B.C. & Vermeirssen, E.L.M. 2000. Distinguishing between female sperm choice versus male sperm competition: a comment on birkhead. *Evolution* **54**: 1050–1052.
- Kolliker, M., Heeb, P., Werner, I., Mateman, A.C., Lessells, C.M. & Richner, H. 1999. Offspring sex ratio is related to male body size in the great tit (*Parus major*). *Behav. Ecol.* **10**: 68–72.
- Komdeur, J., Daan, S., Tinbergen, J. & Mateman, C. 1997. Extreme adaptive modification in sex ratio of the Seychelles warbler's eggs. *Nature (London)* **385**: 522–525.
- Krackow, S. & Tkadlec, E. 2001. Analysis of brood sex ratios: implications of offspring clustering. *Behav. Ecol. Sociobiol.* **50**: 293–301.
- Lessells, C.M. & Boag, P.T. 1987. Unrepeatable repeatabilities: a common mistake. *Auk* **104**: 116–121.
- Madsen, T., Shine, R., Loman, J. & Hakansson, T. 1992. Why do female adders copulate so frequently? *Nature* **355**: 440–442.
- Mange, A. 1970. Possible nonrandom utilization of X- and Y-bearing sperm in *Drosophila melanogaster*. *Genetics* **65**: 95–106.
- Olsson, M., Madsen, T., Shine, R., Gullberg, A. & Tegelstrom, H. 1994. Rewards of promiscuity. *Nature* **372**: 230.
- Olsson, M., Shine, R., Gullberg, A., Madsen, T. & Tegelström, H. 1996. Female lizards control paternity of their offspring by selective use of sperm. *Nature* **383**: 585.
- Petrie, M. 1994. Improved growth and survival of offspring of peacocks with more elaborate trains. *Nature* **371**: 598–599.
- Radford, A.N. & Blakey, J.K. 2000. Intensity of nest defense is related to offspring sex ratio in the great tit *Parus major*. *Proc. R. Soc. Lond. B* **267**: 535–538.
- Reeder, T.W. & Wiens, J.J. 1996. Evolution of the lizard family Phrynosomatidae as inferred from diverse types of data. *Herpes Monographs* **10**: 43–84.
- Rice, W.R. & Chippindale, A.K. 2001. Intersexual ontogenetic conflict. *J. Evol. Biol.* **14**: 685–693.
- Sheldon, B.C., Andersson, S., Griffith, S.C., Ornborg, J. & Sendecka, J. 1999. Ultraviolet colour variation influences blue tit sex ratios. *Nature (London)* **402**: 874–877.
- Sinervo, B. & Calsbeek, R. Ontogenetic conflict between the sexes driven by sexual selection and antagonistic natural selection. *Nature* (submitted).

- Sinervo, B. & Lively, C.M. 1996. The rock-paper-scissors game and the evolution of alternative male reproductive strategies. *Nature* **380**: 240–243.
- Sinervo, B., Svensson, E. & Comendant, T. 2000. Density cycles and an offspring quantity and quality game driven by natural selection. *Nature* **406**: 985–988.
- Svensson, E. & Nilsson, J.-A. 1996. Mate quality affects offspring sex ratio in blue tits. *Proc. R. Soc. Lond. B* **263**: 357–361.
- Tregenza, T. & Wedell, N. 2002. Polyandrous females avoid costs of inbreeding. *Nature (London)* **415**: 71–73.
- Vacquier, V.D. 1995. Evolution of gamete recognition. *Science (Washington, DC)* **285**: 1995–1998.
- West, S.A. & Sheldon, B.C. 2002. Constraints in the evolution of sex ratio adjustment. *Science (Washington, DC)* **295**: 1685–1688.
- Wilkinson, G.S. & Reillo, P.R. 1994. Female choice response to artificial selection on a exaggerated male trait in a stalk-eyed fly. *Proc. R. Soc. Lond. B* **255**: 1–6.
- Wilkinson, G.S., Presgraves, D.C. & Crymes, L. 1998. Male eye span in stalk-eyed flies indicates genetic quality by meiotic drive suppression. *Nature* **391**: 276–278.
- Yasui, Y. 1997. A “good-sperm” model can explain the evolution of costly multiple mating by females. *Am. Nat.* **149**: 573–584.
- Zamudio, K. & Sinervo, B. 2000. Polygyny, mate-guarding, and posthumous fertilizations as alternative male strategies. *Proc. Natl. Acad. Sci. USA* **97**: 14427–14432.

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