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Maternal condition and previous reproduction interact to affect offspring sex in a wild mammal

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Trivers and Willard proposed that offspring sex ratio should vary with maternal condition when condition, meant as maternal capacity to care, has different fitness consequences for sons and daughters. In polygynous and dimorphic species, mothers in good condition should preferentially produce sons, whereas mothers in poor condition should produce more daughters. Despite its logical appeal, support for this hypothesis has been inconsistent. Sex-ratio variation may be influenced by additional factors, such as environmental conditions and previous reproduction, which are often ignored in empirical studies. We analysed 39 years of data on bighorn sheep (*Ovis canadensis*) that fit all the assumptions of the Trivers–Willard hypothesis. Production of sons increased with maternal condition only for mothers that weaned a son the previous year. This relationship likely reflects a mother's ability to bear the higher reproductive costs of sons. The interaction between maternal condition and previous weaning success on the probability of producing a son was independent of the positive effect of paternal reproductive success. Maternal and paternal effects accounted for similar proportions of the variance in offspring sex. Maternal reproductive history should be considered in addition to current condition in studies of sex allocation.

1. Introduction

Sex-allocation theory attempts to explain variation in offspring sex ratio at different levels, from individuals and populations to species. Most research on individual sex allocation in vertebrates is inspired by the Trivers–Willard hypothesis (TWH), which proposes that the sex of offspring should vary with maternal condition when it influences the fitness of sons and daughters differently [1]. Trivers–Willard effects are predicted when (i) maternal condition correlates with offspring condition at the end of maternal care, (ii) differences in condition between offspring at the end of care persist to adulthood and (iii) differences in adult condition have a greater impact on reproductive success of one sex. For polygynous and dimorphic species, the theory predicts that females in good condition should preferentially produce sons, whereas females in poor condition should produce more daughters [1]. However, empirical results are mixed [2–6]. Many species that fit all the assumptions of the TWH do not fit its prediction [2].

Heterogeneous measures of condition may explain inconsistencies in tests of the TWH. Studies measuring maternal condition near conception rather than after birth show stronger support for the hypothesis [3,4]. Furthermore, relative changes in maternal condition seem to better predict offspring sex than absolute values [7]. It is important to note that Trivers and Willard used 'condition' to refer to maternal capacity to care. Environmental conditions and previous reproduction may thus confound the relationship between behavioural or

morphological measures of condition and sex ratio by influencing maternal ability to care. In red deer (*Cervus elaphus*), a positive relationship between maternal dominance and production of sons disappeared at high density, but the adaptive significance of this change remains unclear [8]. To our knowledge, no study has explored whether Trivers–Willard effects depend on reproductive history.

We tested whether environmental conditions near conception and previous weaning success (PWS) affected the relationship between relative condition, measured as yearly changes in mass and offspring sex in female bighorn sheep (*Ovis canadensis*). This species satisfies all the assumptions of the TWH but has repeatedly been found not to support its predictions [9–11].

2. Material and methods

(a) Study population and data

Data were collected from 1975 to 2014 at Ram Mountain, about 30 km east of the Rockies in Alberta, Canada. From late May to late September, sheep were captured in a trap baited with salt. Females can produce one lamb per year from 2 years of age onwards. Lambs were sexed at first capture, aged from one week to three months. Maternity was accurately determined from field observations of suckling behaviour.

We analysed offspring sex of females aged ≥5 years (see electronic supplementary material, table S1 for female age distribution) to obtain a measure of condition relatively independent of female age. We measured maternal condition as a female's mass adjusted to 15 September each year, about two months before conception, minus her average mid-September mass from age 4 onwards. Offspring sex and maternal condition were measured at age x and age $x - 1$, respectively. By age 4, females reach about 95% of asymptotic mass [12]. Females can gain mass until 6 years and senescence-related mass loss begins at 11–12 years [12]. To ensure that our analyses were not biased because of late growth and senescence, we re-ran them considering only females aged 7–13 years. Almost all females (95%) were weighed two to seven times each summer from age 4. Mass adjustments to mid-September are described elsewhere [13]. Our measure of condition provides a direct test of the TWH because relative rather than absolute values of mass correlate with reproductive potential [14].

Because sons are costlier than daughters [15], we distinguished three levels of PWS: no lamb weaned, weaned a female or weaned a male. We used yearly average mass of yearling females in mid-September as an integrator of environmental conditions before conception [16]. Electronic supplementary material, figure S1 summarizes when each variable was measured.

(b) Statistical analyses

We ran generalized linear mixed models (GLMMs) with a binomial distribution using the 'lme4' library [17] in R v. 3.1.2 (http://www.r-project.org) to test whether the probability to produce a son was influenced by maternal condition, PWS, environmental conditions and their interactions. Mother identity and year of lamb birth were fitted as random effects.

Both maternal age and paternal reproductive success (the log-transformed percentage of lambs sired by the father that year) influence offspring sex in this population [10,18]. These two variables were included in the final GLMM to check that the results obtained in the above analyses did not change. Paternity is available for most lambs only since 1988. Father identity was entered as an additional random effect in models including paternal effects.

Table 1. Generalized linear mixed model of the probability of producing a son based on 560 observations of 165 female bighorn sheep. The analysis of deviance table reports the effects of maternal condition, previous weaning success (PWS), environmental conditions (ENV) and their interactions, denoted by ×. Parameter values are given for the final model, excluding non-significant terms.

analysis of deviance	χ^2	d.f.	*p*-value
maternal condition × PWS × ENV	1.55	2	0.47
maternal condition × ENV	1.24	1	0.31
PWS × ENV	0.78	2	0.70
maternal condition × PWS	12.99	2	0.002
ENV	0.75	1	0.40
final model	**estimate**	**s.e.**	**p-value**
intercept	−0.213	0.178	0.23
PWS[a]			
no lamb weaned	0.112	0.224	0.62
weaned a female	0.371	0.227	0.10
maternal condition	1.085	0.343	0.001
no lamb weaned × maternal condition	−1.549	0.465	<0.001
weaned a female × maternal condition	−1.120	0.467	0.016

[a]Females that weaned a male were considered as reference.

We tested successively interaction terms and, if these were not significant, the main effects of variables using likelihood-ratio tests based on bootstrapping of 400 replicates (R script available in electronic supplementary material, S3). Model simplification proceeded until only terms significant at the $p < 0.05$ level remained in the model. We reported standardized regression coefficients for the final model by first centring and then dividing all continuous variables by 2 s.d., to compare effect sizes [19]. We also calculated total variance explained by random and fixed effects [20]. Generalized variance-inflation factors (GVIFs) were calculated for each predictor and full model with the function 'vif' of package 'car'. Collinearity was not a problem because none of the GVIFs exceeded 4.5 [21].

3. Results

Sex ratio did not differ from 0.5 (267 sons, 293 daughters; $\chi^2_{(1)} = 1.12$, $p = 0.29$). A significant interaction between maternal condition and PWS influenced the probability of producing a son, independently of environmental conditions (table 1). After weaning a son, sex ratio varied from a strong daughter bias for mothers in poor condition to a bias towards sons for mothers in good condition (figure 1*a*). Production of sons appeared to decrease with condition when mothers failed to wean a lamb the previous year, but this pattern was not statistically significant (figure 1*c*). Neither environmental conditions nor their interaction with maternal condition influenced the probability of producing a son (table 1). Restricting
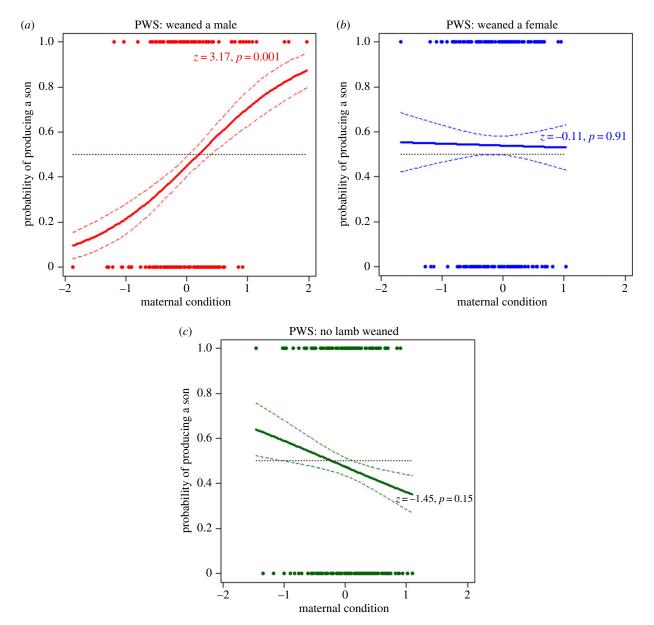
Figure 1. Probability of producing a son according to maternal condition and previous weaning success (PWS): (*a*) weaned a male, (*b*) weaned a female and (*c*) no lamb weaned. We obtained *z* statistic of whether the slope was statistically equivalent to zero by changing the reference level of PWS in the final model (table 1). Solid and dashed lines represent model predictions $\pm$ s.e. These relationships are robust to exclusion of extreme values of maternal condition ($n = 4$, electronic supplementary material, figure S2). The straight dotted lines show a probability of 0.5.

the analysis to females aged 7–13 years yielded similar results (electronic supplementary material, table S2).

Re-running the final model (table 1) with paternal reproductive success or maternal age as additional explanatory variable did not alter the conclusions (electronic supplementary material, tables S3 and S4). The probability of producing a son was positively influenced by paternal success (slope $= 0.886 \pm 0.281$, $p = 0.002$), but not by maternal age (slope $= 0.057 \pm 0.190$, $p = 0.77$). The model with paternal success and the interaction between maternal condition and PWS (electronic supplementary material, table S4) explained 11.9% of the variation in offspring sex, of which 9.9% was explained by the fixed effects. Maternal and paternal fixed effects taken separately accounted for 4.6% and 4.4%, respectively, of the model variance.

4. Discussion

Tests of the TWH in mammals have produced inconsistent results [2,4]. Understanding the causes of such variation is

a challenge for sex-allocation theory. Schindler and colleagues recently suggested that sexual differences in lifetime reproductive value explain the mismatch between TWH predictions and observations [11]. After accounting for sex differences in demography, however, their model still predicted a Trivers–Willard effect in bighorn sheep. We found that production of sons increased with maternal condition only for mothers that weaned a son the previous year.

A correlation between offspring sex and maternal condition may arise simply as a consequence of higher male mortality during early life [8]. We did not know sex ratio at birth, but neonatal mortality was relatively low (17%) compared with subsequent juvenile mortality (45%) with no evidence that it was sex-specific [18]. It is also unlikely that our results were due to sex-specific neonatal mortality since the relationship between sex ratio and maternal condition after weaning a son was symmetrical around 0.5 [3,7]. Furthermore, the hypothesis invoking male-biased early mortality predicts excess daughters following years of poor environmental conditions [22], which was not the case.

Our findings may instead reflect adaptive manipulation of offspring sex. In polygynous dimorphic ungulates, higher energetic demands of sons over daughters can lead to higher costs in terms of future reproduction and survival for mothers that raised sons [2]. A previous study reported that bighorn females avoided producing sons in consecutive years [15]. Our analyses reveal that this is true only when females are in poor condition, likely because the costs of reproduction are highest. Females in good condition after weaning a son likely acquired a large amount of resources, so it would be advantageous for them to bias care towards sons. A combination of these two effects can explain why only mothers that weaned a son the previous year behave as predicted by Trivers and Willard.

Generally, sex-ratio studies of vertebrates yield small effect sizes [5]. A recent meta-analysis reported that paternal attractiveness explained less than 1% of variation in offspring sex ratio [23]. About 10% of variation in offspring sex of bighorn sheep was explained by an equal contribution of maternal and paternal effects. Our results emphasize the importance of considering maternal reproductive history in addition to condition to predict sex ratio, because the product of these two variables better reflects a mother's capacity to care.

References

1. Trivers RL, Willard DE. 1973 Natural selection of parental ability to vary the sex ratio of offspring. *Science* **179**, 90–92. (doi:10.1126/science.179.4068.90)

2. Hewison AM, Gaillard JM. 1999 Successful sons or advantaged daughters? The Trivers–Willard model and sex-biased maternal investment in ungulates. *Trends Ecol. Evol.* **14**, 229–234. (doi:10.1016/S0169-5347(99)01592-X)

3. Sheldon BC, West SA. 2004 Maternal dominance, maternal condition, and offspring sex ratio in ungulate mammals. *Am. Nat.* **163**, 40–54. (doi:10.1086/381003)

4. Cameron EZ. 2004 Facultative adjustment of mammalian sex ratios in support of the Trivers–Willard hypothesis: evidence for a mechanism. *Proc. R. Soc. Lond. B* **271**, 1723–1728. (doi:10.1098/rspb.2004.2773)

5. West SA. 2009 *Sex allocation*. Princeton, NJ: Princeton University Press.

6. Komdeur J. 2012 Sex allocation. In *Evolution of parental care* (eds NJ Royle, PT Smiseth, M Kölliker), pp. 171–188. Oxford, UK: Oxford University Press.

7. Cameron EZ, Linklater WL. 2007 Extreme sex ratio variation in relation to change in condition around conception. *Biol. Lett.* **3**, 395–397. (doi:10.1098/rsbl.2007.0089)

8. Kruuk LEB, Clutton-Brock TH, Albon SD, Pemberton JM, Guinness F. 1999 Population density affects sex ratio variation in red deer. *Nature* **399**, 459–461. (doi:10.1038/20917)

9. Blanchard P, Festa-Bianchet M, Gaillard JM, Jorgenson JT. 2004 Maternal condition and offspring sex ratio in polygynous ungulates: a case study of bighorn sheep. *Behav. Ecol.* **16**, 274–279. (doi:10.1093/beheco/arh153)

10. Martin JGA, Festa-Bianchet M. 2011 Sex ratio bias and reproductive strategies: what sex to produce when? *Ecology* **92**, 441–449. (doi:10.1890/09-2413.1)

11. Schindler S, Gaillard JM, Grüning A, Neuhaus P, Traill LW, Tuljapurkar S, Coulson T. 2015 Sex-specific demography and generalization of the Trivers–Willard theory. *Nature* **526**, 249–252. (doi:10.1038/nature14968)

12. Nussey D, Coulson T, Delorme D, Clutton-Brock T, Pemberton J, Festa-Bianchet M, Gaillard JM. 2011 Patterns of body mass senescence and selective disappearance differ across three species of free-living ungulates. *Ecology* **92**, 1936–1947. (doi:10.1890/11-0308.1)

13. Martin JGA, Pelletier F. 2011 Measuring growth patterns in the field: effects of sampling regime and methods on standardized estimates. *Can. J. Zool.* **89**, 529–537. (doi:10.1139/z11-018)

14. Festa-Bianchet M. 1998 Condition-dependent reproductive success in bighorn ewes. *Ecol. Lett.* **1**, 91–94. (doi:10.1046/j.1461-0248.1998.00023.x)

15. Bérubé CH, Festa-Bianchet M, Jorgenson JT. 1996 Reproductive costs of sons and daughters in Rocky Mountain bighorn sheep. *Behav. Ecol.* **7**, 60–68. (doi:10.1093/beheco/7.1.60)

16. Festa-Bianchet M, Coltman DW, Turelli L, Jorgenson JT. 2004 Relative allocation to horn and body growth in bighorn rams varies with resource availability. *Behav. Ecol.* **15**, 305–312. (doi:10.1093/beheco/arh014)

17. Bates D, Maechler M, Bolker B, Walker S. 2015 Fitting linear mixed-effects models using lme4. *J. Stat. Softw.* **67**, 1–48. (doi:10.18637/jss.v067.i01)

18. Douhard M, Festa-Bianchet M, Coltman DW, Pelletier F. 2016 Paternal reproductive success drives sex allocation in a wild mammal. *Evolution* **70**, 358–368. (doi:10.1111/evo.12860)

19. Gelman A. 2008 Scaling regression inputs by dividing by two standard deviations. *Stat. Med.* **27**, 2865–2873. (doi:10.1002/sim.3107)

20. Nakagawa S, Schielzeth H. 2013 A general and simple method for obtaining R^2 from generalized linear mixed-effects models. *Methods Ecol. Evol.* **4**, 133–142. (doi:10.1111/j.2041-210x.2012.00261.x)

21. Dormann CF et al. 2013 Collinearity: a review of methods to deal with it and a simulation study evaluating their performance. *Ecography* **36**, 27–46. (doi:10.1111/j.1600-0587.2012.07348.x)

22. Lindström J, Coulson T, Kruuk L, Forchhammer MC, Coltman DW, Clutton-Brock T. 2002 Sex-ratio variation in Soay sheep. *Behav. Ecol. Sociobiol.* **53**, 25–30. (doi:10.1007/s00265-002-0545-4)

23. Booksmythe I, Mautz B, Davis J, Nakagawa S, Jennions MD. In press. Facultative adjustment of the offspring sex ratio and male attractiveness: a systematic review and meta-analysis. *Biol. Rev.* (doi:10.1111/brv.12220)

24. Douhard M, Festa-Bianchet M, Pelletier F. 2016 Data from: Maternal condition and previous reproduction interact to affect offspring sex in a wild mammal. Dryad Digital Repository: http://dx.doi.org/10.5061/dryad.q7c24.