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Sex allocation theory aids species conservation

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Supplementary feeding is often a key tool in the intensive management of captive and threatened species. Although it can increase such parameters as breeding frequency and individual survival, supplementary feeding may produce undesirable side effects that increase overall extinction risk. Recent attempts to increase breeding frequency and success in the kakapo *Strigops habroptilus* using supplementary feeding inadvertently resulted in highly male-biased chick sex ratios. Here, we describe how the inclusion of sex allocation theory has remedied this conservation dilemma. Our study is the first to manipulate chick sex ratios in an endangered species by altering maternal condition and highlights the importance of incorporating evolutionary theory into modern conservation practice.

Keywords: evolutionary theory; sex allocation; conservation; supplementary feeding

1. INTRODUCTION

Evolutionary theory predicts that females should manipulate the sex of their offspring in response to characteristics of the rearing environment (Trivers & Willard 1973). While still controversial (West & Sheldon 2002; Ewen *et al.* 2004), compelling examples of sex allocation exist, particularly among birds (Heinsohn *et al.* 1997; Komdeur *et al.* 1997). Recent avian studies have highlighted the influence of maternal condition on brood sex ratios (Nager *et al.* 1999; Whittingham & Dunn 2000). This relationship has important implications for the use of supplementary feeding, especially in conservation, where it is often used as a key tool in the intensive management of threatened species (Castro *et al.* 2003).

The kakapo (*Strigops habroptilus*, Gray 1845), a critically endangered New Zealand parrot, has been subject to supplementary feeding since 1989: feeding improves female condition and egg and chick survival (Elliott *et al.* 2001). Although supplementary feeding has been adaptively managed to avoid short-term negative effects, such as female obesity (Elliott *et al.* 2001), little consideration was given to long-term effects until Tella (2001) suggested that increased maternal condition associated with supplementary feeding might influence sex ratios in kakapo. As predicted from evolutionary theory and kakapo life-history traits, such as lek mating (Clout *et al.* 2002), skewed male reproductive success based on body size (Clout *et al.* 2002; Miller *et al.* 2003) and larger faster growing male nestlings than females (Elliott *et al.* 2001), analysis of sex allocation in kakapo revealed that fed females produced more sons than daughters (i.e. 70% of offspring were sons; Clout *et al.* 2002).

Male-biased production in an already male-biased population (currently 45 males : 38 females) represents a critical challenge to kakapo recovery by prolonging species recovery and the risk of extinction. The solution to this dilemma, though, is not simply to cease supplementary feeding, because feeding plays a significant role in kakapo recovery. Female kakapo do not breed unless they are above a threshold weight of 1.5 kg (Elliott *et al.* 2001), hence supplementary feeding can ensure all females are above the threshold weight prior to the breeding season and hence, potentially stimulate breeding.

Just as evolutionary theory identified the quandary facing kakapo conservators, it also provides a solution through altering maternal condition (Wedekind 2002). Here, we report a new feeding regime for kakapo that achieved our two distinct objectives for supplementary feeding in kakapo recovery. First, it raised all adult females above the 1.5 kg breeding threshold and thereby potentially stimulated breeding. Second, it removed male-biased sex allocation by ensuring that females do not attain the body condition associated with male-biased brood sex ratios (Clout *et al.* 2002). This manipulation of chick sex ratios in an endangered species by controlling maternal condition is a first for conservation science and highlights the potential for achieving positive conservation outcomes through the application of evolutionary theory.

2. MATERIAL AND METHODS

The new feeding regime was initiated in winter 2001, when female kakapo were at their minimum body condition (Elliott *et al.* 2001). In contrast to previous regimes (Clout *et al.* 2002), in which all fed females were provisioned with *ad libitum* food, only females below a *predicted* December weight of 1.5 kg (estimated using winter weights corrected for month: Elliott *et al.* 2001) were given *ad libitum* food in the months before commencement of mating in Mid-January 2002 (i.e. *ad libitum* fed females). Females above the predicted December weight threshold of 1.5 kg were not given *ad libitum* food to avoid raising their body condition to that previously associated with male-bias sex allocation (i.e. restricted fed females). Restricted fed females, however, were minimally provisioned to maintain their interest in the food dispensers. Following mating, all females were provided with *ad libitum* food to increase egg and chick survival.

Kakapo breed in synchrony with heavy fruiting of native trees (Clout *et al.* 2002) laying 2.53 ± 0.01 eggs ($n = 54$ broods; Eason *et al.* 2006). The 2002 season was a good breeding year for kakapo with 20 of 21 adult females laying (Eason *et al.* 2006): 67 eggs were produced, of which 42 were fertile, 26 hatched and 24 fledged. We determined brood sex ratios by sexing all nestlings and deceased embryos using PCR amplification conditions in Robertson *et al.* (2000). To effectively reduce sexing error, all male genotypes were verified with primers CHD2550/2718 (Fridolfsson & Ellegren 1999; Robertson & Gemmell in press).

We examined the effect of our new feeding regime on female weight with t tests using StatView (SAS Institute, Inc.) and brood sex ratios with binomial logistic regression following Wilson & Hardy (2002) using the GLM function in S-Plus 4.5 (Mathsoft, Inc.). Statistical validity ($\alpha = 0.05$) was tested with a *post hoc* power ($1-\beta$) analysis using Gpower (Erdfelder *et al.* 1996). In addition, we calculated the difference in proportion of males for *ad libitum* fed and restricted fed female broods using a randomization test

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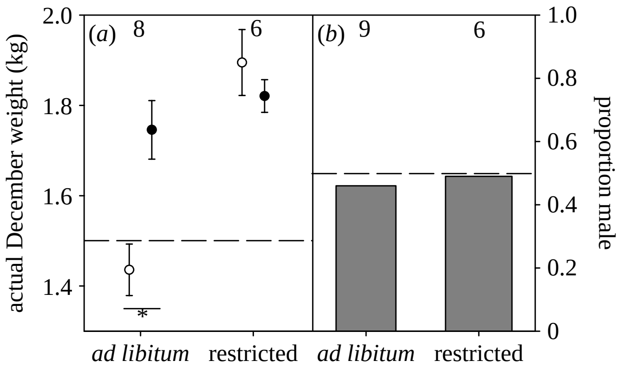

Figure 1. (*a*) Breeding weight and (*b*) brood sex ratios of female kakapo in relation to *ad libitum* and restricted supplementary feeding. Open and closed circles denote the mean predicted and actual December weights of female kakapo, respectively. Grey bars represent mean brood sex ratios of kakapo clutches. Error bars are standard errors. Values are (*a*) number of females and (*b*) number of broods. The dashed lines represent the 1.5 kg breeding weight threshold and a 50 : 50 sex ratio, respectively. $^*p < 0.05$.

(Sokal & Rohlf 1995) based on the sexes of chicks in each brood ($n = 100$ randomizations). Values were ranked to determine the percentile of the observed value. As in Clout *et al.* (2002), we found no sex-bias among first laid eggs: three ♂ eggs in seven *ad libitum* fed females and two ♂ eggs in five restricted fed females (incomplete hatch order data limited further analysis). All reported values are mean ± standard error unless otherwise stated.

3. RESULTS

Our new feeding regime raised all female weights (i.e. *actual* December weights) above the 1.5 kg breeding threshold (figure 1): actual weights of *ad libitum* females were significantly greater than predicted weights ($t_7 = -5.197$, $p = 0.001$) and similar to the actual weights of females on the restricted diet ($t_{12} = -0.91$, $p = 0.38$). Our feeding regime appears to have also controlled the male-biased sex allocation associated with previous regimes. Molecular sexing showed that *ad libitum* fed females produced a total of nine male and ten female eggs, whereas restricted fed females produced seven male and nine female eggs. Brood sex ratios of *ad libitum* fed and restricted fed females were close to parity (figure 1). Furthermore, binomial logistic regression indicated that our new feeding regime had no effect on brood sex ratios (all clutches, $F_{1,13} = 0.1769$, $p = 0.68$, power = 0.89; clutches containing unsexed fertile eggs removed, $F_{1,9} = 0.098$, $p = 0.76$, power = 0.89). The observed difference in proportion of males from *ad libitum* fed and restricted fed female broods was in the 49th percentile of randomization values, adding further statistical evidence that our new supplementary feeding regime produced sex ratios close to parity and removed the male bias brood sex ratios noted previously.

4. DISCUSSION

Our optimized feeding regime was an outstanding success for kakapo species recovery fulfilling both our conservation objectives. First, targeted feeding raised all predicted-underweight female kakapo above the 1.5 kg breeding threshold, thereby potentially stimulating breeding. Indeed, all but one adult female kakapo bred in the 2002 season, which might in part be consequence of all females being above the 1.5 kg breeding threshold. The only female not to breed, Jane, has never bred since her discovery on Stewart Island in 1989. Second, in line with expectations from sex allocation theory, the new supplementary feeding regime removed the male bias in offspring production previously noted in kakapo (Clout *et al.* 2002) by limiting female body condition below the high level that induced an over production of sons (e.g. actual breeding weights, 1.98 ± 0.23 kg (±s.d.), range: 1.69–2.33 kg, $n = 6$ supplementary fed females prior to 2002; Elliott *et al.* 2001). As such, our study is the first to manipulate chick sex ratios in an endangered species by altering maternal condition (Wedekind 2002) and highlights the importance of incorporating evolutionary theory into modern conservation to optimize management practices.

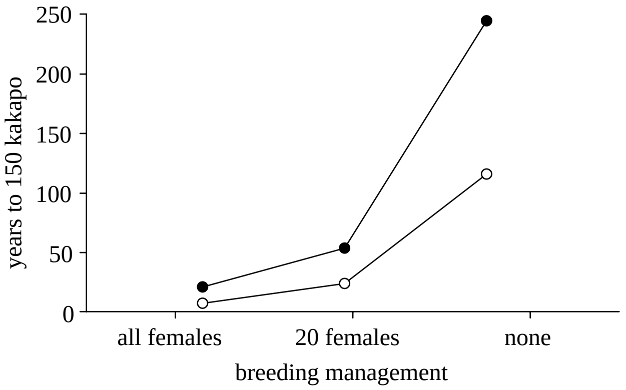

Figure 2. Predicted impact of male-biased brood sex ratios on kakapo recovery under various levels of breeding management (all females, all breeding females intensively managed; 20 females, only 20 breeding females are managed; none, no management of breeding females) using a stochastic model and Monte Carlo simulation of kakapo population trajectory (Elliott 2006). Open circles are 50 : 50 brood sex ratios and closed circles are 70 : 30 male-biased brood sex ratios.

Other factors might have produced the 50 : 50 sex ratio, such as stochastic variation in brood sex ratios independent of the influence of supplementary feeding or different environmental conditions between years. Between-year effects might be weak in kakapo, as females above the 1.5 kg weight threshold only breed in years of abundant resources (i.e. years with heavy fruiting of native trees; Clout *et al.* 2002). Given abundant resources, females might be expected to show a male bias (e.g. Komdeur *et al.* 1997), not a 50 : 50 sex ratio, adding further support to our assertion that we have manipulated kakapo sex ratios. The potential for chance and between year effects means we can only be completely confident that our new feeding regime has removed the male-bias if we conduct controlled experiments to better understand sex allocation in kakapo. Such an approach is currently not feasible due to the critically endangered status of kakapo, which is why we took our precautionary, adaptive approach to management of the sex bias. In the interim, monitoring of brood sex ratios should indicate if the new feeding regime continues to limit sex biased production in kakapo.

The male bias in kakapo productivity presented a considerable challenge to kakapo's recovery. The observed overproduction of sons (Clout *et al.* 2002) would have hindered kakapo population growth by between 12 and 109 years depending on the level of breeding management (figure 2; Elliott 2006), thereby elevating the risk of extinction for this species. By using lessons learned from more abundant species and an understanding of the life-history traits of kakapo that lead to a large fitness differential in the production of sons and daughters, we successfully instigated a management programme that has removed the bias in offspring sex ratios.

Our deliberate manipulation of kakapo sex ratios, based on predictions from sex allocation theory, is a unique example of the successful application of evolutionary theory to achieve a vital conservation objective. As such, our study highlights the significance of considering the impact of maternal condition in conservation practice (Wedekind 2002) and should have important implications for the conservation and captive management of the ever-increasing number of endangered species.

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Castro, I., Brunton, D. H., Mason, K. M., Ebert, B. & Griffiths, R. 2003 Life history traits and food supplementation affect productivity in a translocated population of the endangered hihi (stitchbird *Notiomystis cincta*). *Biol. Conserv.* **114**, 271–280. (doi:10.1016/S0006-3207(03)00046-6)

Clout, M. N., Elliott, G. P. & Robertson, B. C. 2002 Effects of supplementary feeding on the offspring sex ratio of kakapo: a dilemma for the conservation of a polygynous parrot. *Biol. Conserv.* **107**, 13–18. (doi:10.1016/S0006-3207(01)00267-1)

Eason, D. K., Elliott, G. P., Harper, G. A. & Moorhouse, R. J. 2006 Breeding biology of kakapo (*Strigops habroptilus*) on offshore island sanctuaries, 1990–2002. *Notornis* **53**, 27–36.

Elliott, G. P. 2006 A simulation of the future of kakapo. *Notornis* **53**, 164–172.

Elliott, G. P., Merton, D. V. & Jansen, P. W. 2001 Intensive management of a critically endangered species: the kakapo. *Biol. Conserv.* **99**, 121–133. (doi:10.1016/S0006-3207(00)00191-9)

Erdfelder, E., Faul, F. & Buchner, A. 1996 GPOWER: a general power analysis program. *Behav. Res. Meth. Instrum. Comput.* **28**, 1–11.

Ewen, J. G., Cassey, P. & Moller, A. P. 2004 Facultative primary sex ratio variation: a lack of evidence in birds? *Proc. R. Soc. B* **271**, 1277–1282. (doi:10.1098/rspb.2004.2735)

Fridolfsson, A. K. & Ellegren, H. 1999 A simple and universal method for molecular sexing of non-ratite birds. *J. Avian Biol.* **30**, 116–121.

Heinsohn, R., Legge, S. & Barry, S. 1997 Extreme bias in sex allocation in *Eclectus* parrots. *Proc. R. Soc. B* **264**, 1325–1329. (doi:10.1098/rspb.1997.0183)

Komdeur, J., Daan, S., Tinbergen, J. & Mateman, C. 1997 Extreme adaptive modification in sex ratio of the Seychelles warbler's eggs. *Nature* **385**, 522–525. (doi:10.1038/385522a0)

Miller, H. C., Lambert, D. M., Millar, C. D., Robertson, B. C. & Minot, E. O. 2003 Minisatellite DNA profiling detects lineages and parentage in the endangered kakapo (*Strigops habroptilus*) despite low microsatellite DNA variation. *Conserv. Genet.* **4**, 265–274. (doi:10.1023/A:1024037601708)

Nager, R. G., Monaghan, P., Griffiths, R., Houston, D. C. & Dawson, R. 1999 Experimental demonstration that offspring sex ratio varies with maternal condition. *Proc. Natl Acad. Sci. USA* **96**, 570–573. (doi:10.1073/pnas.96.2.570)

Robertson, B. C. & Gemmell, N. J. In press. PCR-based sexing in conservation biology: wrong answers from an accurate methodology? *Conserv. Genet.* (doi:10.1007/s10592-005-9105-6)

Robertson, B. C., Millar, C. D., Minot, E. O., Merton, D. V. & Lambert, D. M. 2000 Sexing the critically endangered kakapo *Strigops habroptilus*. *Emu* **100**, 336–339. (doi:10.1071/MU00056)

Sokal, R. R. & Rohlf, F. J. 1995 *Biometry: the principles and practice of statistics in biological research*. New York: W.H. Freeman & Co.

Tella, J. L. 2001 Sex-ratio theory in conservation biology. *Trends Ecol. Evol.* **16**, 76–77. (doi:10.1016/S0169-5347(00)02075-9)

Trivers, R. L. & Willard, D. E. 1973 Natural selection of parental ability to vary the sex ratio of offspring. *Science* **179**, 90–92.

Wedekind, C. 2002 Manipulating sex ratios for conservation: short-term risks and long term benefits. *Anim. Conserv.* **5**, 13–20.

West, S. A. & Sheldon, B. C. 2002 Constraints in the evolution of sex ratio adjustment. *Science* **295**, 1685–1688. (doi:10.1126/science.1069043)

Whittingham, L. A. & Dunn, P. O. 2000 Offspring sex ratios in tree swallows: females in better condition produce more sons. *Mol. Ecol.* **9**, 1123–1129. (doi:10.1046/j.1365-294x.2000.00980.x)

Wilson, K. & Hardy, I. C. W. 2002 Statistical analysis of sex ratios: an introduction. In *Sex ratios: concepts and research methods* (ed. I. C. W. Hardy), pp. 48–92. Cambridge, UK: Cambridge University Press.