Offspring fitness and individual optimization of clutch size

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Within-year variation in clutch size has been claimed to be an adaptation to variation in the individual capacity to raise offspring. We tested this hypothesis by manipulating brood size to one common size, and predicted that if clutch size is individually optimized, then birds with originally large clutches have a higher fitness than birds with originally small clutches. No evidence was found that fitness was related to the original clutch size, and in this population clutch size is thus not related to the parental capacity to raise offspring. However, offspring from larger original clutches recruited better than their nest-mates that came from smaller original clutches. This suggests that early maternal or genetic variation in viability is related to clutch size.

Keywords: life history; clutch size optimization; recruitment; *Parus major*; cross-fostering

1. INTRODUCTION

One of the major trade-offs in life-history theory is between the quality and number of offspring (Lessells 1991). Ideally one would express offspring quality in terms of its lifetime fitness, but this measurement is often difficult to obtain. Most evidence for the existence of the trade-off comes from studies in which clutch size is manipulated and some physical features of the offspring are measured, such as mass or size at fledging or independence (Lack 1947; Perrins & Moss 1975; Dijkstra *et al.* 1989; Tinbergen & Daan 1990). Size and fledging mass have been shown to be important determinants of recruitment probability in various bird species (Perrins 1965; Garnett 1981; Tinbergen & Boerlijst 1990; Magrath 1991; Verboven & Visser 1998). There might, however, be additional differences between individual chicks that are not reflected in their body mass but do determine their survival probability (see Verhulst 1994; Saino *et al.* 1997). These differences might be either genetically or environmentally determined. If variation in chick quality is related to a variable like clutch size, this has profound implications for the design of brood size manipulation experiments, because these experiments assume that offspring from different broods are on average of equal initial quality.

Quality differences between offspring from different parents can be investigated in experiments in which chicks from different parents grow up under the same conditions. These experiments test for genetic differences and early environmental effects, but do not distinguish between them. Genetic variation in offspring viability is mostly unexpected, because strong selection will wipe out all genetic variation for such a trait in a short time (Fisher 1930; Gustafsson 1986). However, Norris (1993)

suggested that in juvenile great tit males there is genetic variance in viability, which is associated with the size of a sexually selected character. An alternative to genetic variation in offspring quality is that egg-laying females alter egg composition, including hormone levels (Schwabl 1993; Schwabl *et al.* 1997) or antibodies (Heeb *et al.* 1998), in relation to internal and/or environmental circumstances. The observation that yolk testosterone levels increase during the laying sequence (Schwabl 1993), might result in a positive relationship between average hormone levels per nest and clutch size. These data imply that parents not only decide how many eggs they lay in a brood, but also simultaneously might decide on the quality of each of the eggs.

If offspring originating from clutches of different sizes do differ in viability, this will have consequences for the interpretation of brood size manipulation experiments. Brood size experiments generally allocate broods, but not the component chicks, randomly to different experimental treatments. If chicks differ in initial quality this will mean that the experiments miss an important part of the clutch size decision. Brood size experiments mostly ignore the variation in offspring quality between different broods, or do cross-foster but do not test explicitly for variation in offspring quality (but, see Gebhardt-Henrich & van Noordwijk 1991, 1994). In this study, we equalized brood sizes to one common brood size by exchanging at least half of a brood with a different nest. In this paper, we test whether the clutch size of the biological parents of cross-fostered chicks is important in determining the chicks' survival prospects. Furthermore, we consider whether parents with an originally larger clutch were better at raising offspring than parents with smaller clutches. The experiment is based upon earlier experimental work in which individual optimization of clutch size could not be shown (Verhulst 1994; Both 1998), despite earlier claims of individual optimization in the

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same species (Perrins & Moss 1975; Pettifor *et al.* 1988; Tinbergen & Daan 1990).

2. METHODS

Brood size manipulations are necessary to separate the fitness consequences of variation in brood size and parental quality (Perrins & Moss 1975; Van Noordwijk & De Jong 1986; Lessells 1991). Most brood size experiments compare reduced and enlarged broods of equal initial size to test whether deviations from the actual brood size reduce fitness, as predicted by the individual optimization hypothesis (e.g. Perrins & Moss 1975; Drent & Daan 1980; Högstedt 1980; Pettifor *et al.* 1988; Tinbergen & Daan 1990). In this experiment, we used a slightly different approach to test whether the observed variation in clutch size reflected differences in the parental ability to raise offspring. The experiments were carried out from 1995–1997 on the Buunderkamp in The Netherlands, where data on great tit breeding ecology have been collected since 1983 from the birds breeding in nestboxes (see Drent (1987) for a detailed description of the study area). The experiment was part of a larger experiment in which effects of density and territory size were examined, but no effects of other treatments were found that confound the results presented here. Nest-boxes were checked weekly, and daily at the predicted hatching date. Egg length and breadth were only measured in 1995, and an index of egg size was calculated as $0.5 \times \text{length} \times \text{breadth}^2$. In total, 103 broods were manipulated to contain nine chicks between days 2 and 4 (where day 0 is the day on which the first chick in the nest hatched). Chicks were exchanged between two or three broods that hatched on the same day. Groups of chicks to be exchanged were chosen blind from a brood. Each of the broods in which chicks were exchanged received 3–7 chicks (mean 4.82, s.d. 0.83) from other broods. The size of the group depended on the number of chicks hatched in that particular brood and the number of broods that hatched on the same day. Nests were matched on the basis of the number of chicks hatched, in order to enable manipulation of brood size to nine chicks. In practice, large broods were thus matched with small broods, while intermediate-sized broods were matched with other intermediate-sized broods. On a 'per nest' basis the chicks were divided equally among the matched nests. Although recent work has shown that incubation costs are related to clutch size (Moreno & Sanz 1994; Monaghan & Nager 1997), we have chosen to manipulate in the early chick stage because this reduces the variance in hatching date due to the different incubation patterns of females. If clutch size is adjusted to parental condition or territory quality, females in better circumstances pay the higher incubation cost, and the effect of clutch size on fitness will be conservative.

All chicks were individually marked at the time of manipulation by clipping part of their down, and were ringed with aluminium rings when they were seven days old. Chicks were weighed at the time of manipulation (only in 1995 and 1997), and on day 7 and day 15. Within manipulated nests, the nestling mass at manipulation did not differ between the foster and the original chicks (final nest: $F_{59,63}=2.36$, $p<0.0004$; original vs foster: $F_{1,63}=0.19$, $p=0.66$; the difference between nests is due to variation in chick age at manipulation), while there was no effect of original clutch size on chick mass at this stage (between-nest effect: $F_{59,63}=2.42$, $p<0.0004$; original clutch size: $F_{1,63}=0.20$, $p=0.66$). Sex of the fledglings was estimated from their primary coverts at 15 days old (in all years), and in

1996 and 1997 most nestlings were sexed using RAPD-markers (see Lessells *et al.* 1996; Lessells & Mateman (1998) for detailed methods). When nestlings were sexed by both methods ($n=451$), 82% of the sex estimates from the primary coverts agreed with the sex revealed by the molecular sexing. In the analysis below, the sex obtained using the molecular method was used if available, otherwise the sex estimate based on the wing coverts was used (54% of chicks were sexed with RAPD-markers). Great tits sometimes replace failed clutches or produce second broods after a successful first brood. A chick recruits if it is caught as a breeding bird in the following breeding season. The fitness return from the whole breeding season should be calculated, including the number of recruits from replacement and second broods, because effort expended raising the first brood may affect the success of the subsequent attempts (Tinbergen *et al.* 1985; Tinbergen 1987). Parents were caught in the nestbox on day 7 and ringed with aluminium rings. Parental survival and chick recruitment are based on capture of breeding birds in the following breeding season. Data on effects in the nestling stage are presented for 1995–1997, while survival and recruitment data are only available for families manipulated in 1995 and 1996.

In all cases where the analyses deal with measures before fledging, average values per group of chicks originating from the same brood in each nest were used. In the case of recruitment rates each fledgling was treated as an independent data point, because only then could fledging mass and offspring sex be included in the analysis. Both variables have been shown to influence recruitment (Tinbergen & Boerlijst 1990; Verboven & Visser 1998). In the analysis of the cross-fostering experiment, a factor for final nest is fitted first, in order to compare chicks from different original nests that grow up under the same conditions. Thus we analyse whether variation between chicks from different origins is explained by the clutch size of their biological parents.

Before analysing the differences between chicks we analysed whether parents that laid a larger clutch were also more able parents in raising the same number of offspring. The currency used in this analysis was the number of recruits produced during that breeding season and the survival of the parents to the next year. The number of recruits was based on the chicks that the parents care for, and not on their original chicks which grow up in other broods.

3. RESULTS

(a) *Brood size and fitness*

In the experiment in which all broods were manipulated to contain the same number of chicks, no relationship was found between the number of recruits and the original clutch size of the parents raising the brood (table 1). Neither the nestling survival rate, nor the condition of fledglings expressed as their fledging mass was affected by the size of the original clutch (table 1). Male survival was positively related to the size of the original clutch of the nest (year: $\chi^2_1=0.12$, $p=0.73$; original clutch size: $\chi^2_1=4.96$, $p=0.026$; interaction: $\chi^2_1=0.21$, $p=0.65$; $n=60$), but female survival was not (year: $\chi^2_1=0.67$, $p=0.41$; original clutch size: $\chi^2_1=0.001$, $p=0.97$; interaction: $\chi^2_1=0.58$, $p=0.45$; $n=60$). The representation of parental genes in the next season, i.e. the sum of the number of recruits and the number of parents surviving, was independent of the original clutch

Table 1. *Effects of original clutch size on fitness components in the brood size experiment in which all broods received nine nestlings at days 2–3*

(Fledging mass has been analysed and is normally distributed, nestling survival is binomially distributed (with Williams' correction, Crawley (1993)), and recruitment number is analysed with a Poisson error structure. Number of recruits is from all broods in that season.)

	fledging mass		nestling survival		number of recruits	
	<i>F</i> -value	<i>p</i>	<i>F</i> -value	<i>p</i>	<i>F</i> -value	<i>p</i>
year	$F_{2,89}=11.06$	<0.001	$F_{2,98}=3.61$	0.03	$\chi^2_2=14.7$	<0.001
clutch size	$F_{1,89}=2.27$	0.14	$F_{1,98}=1.46$	0.23	$\chi^2_1=1.07$	0.30
year $\times$ clutch size	$F_{2,87}=0.60$	0.56	$F_{2,96}=1.31$	0.27	$\chi^2_2=0.005$	0.99

size they had laid (Poisson regression, year: $\chi^2_1=6.71$, $p=0.01$; original clutch size: $\chi^2_1=2.21$, $p=0.14$; interaction: $\chi^2_1=0.08$, $p=0.78$). Thus, in the experiment we cannot confirm that parents laying larger clutches are doing so because they are better in raising offspring.

(b) Variation in recruitment related to clutch size

The possibility exists that individual optimization of clutch size could have been masked by offspring differing in viability because their parents adjust offspring quality together with clutch size. The hypothesis that birds that produce larger clutches also have better quality offspring was tested by comparing the offspring from different natal nests that had been cross-fostered to the same nest as part of the manipulation experiment. The recruitment rate of nestlings growing up in the same nest was positively related to the clutch size of their biological parents (figure 1). Fledging mass was not explained by the clutch size of the biological parent (between-final nest effect: $F_{96,108}=2.94$, $p<0.0001$; clutch size of natal nest: $F_{1,108}=0.08$, $p=0.78$), and the effect of original clutch size on recruitment still existed after entering fledging mass in the analysis (see figure 1). No differences within nests were found on recruitment rate between males and females, nor on any interaction between sex, mass and original clutch size (in logistic ANCOVA, all p -values >0.10). Thus, nestlings from larger original clutches were better survivors than from small clutches if they grew up under identical conditions.

One possible variable that might explain the differences in recruitment rate is the egg size. In 1995 mean egg size and clutch size were not correlated ($r=-0.074$, $n=29$), while within nests the egg size of the original parents did not explain variation in recruitment rate (between-final nest effect: $\chi^2_{17}=28.6$; egg size: $\chi^2_1=0.007$, $p=0.93$; $n=144$).

Chicks originating from larger clutches were from broods in which the first eggs were laid earlier than those of their nest-mates originating from smaller clutches. If laying date is related to female condition, then the effect of natal clutch size on recruitment could be an effect of laying date (and thus female condition) rather than of clutch size. In the analysis of recruitment rates, the first egg-laying date of the natal nest was included, but this variable did not explain a significant part of the variance (between-final nest effect: $\chi^2_{50}=75.2$, $p=0.012$; fledging mass: $\chi^2_1=9.19$, $p<0.003$; natal clutch size: $\chi^2_1=8.49$, $p<0.004$; natal laying date: $\chi^2_1=0.18$, $p=0.67$; smaller

Figure 1. The effect of the natal clutch size on the mean recruitment averaged per brood of a given size. The cross-fostering experiment revealed that chicks from larger original clutches recruited better than their nest-mates from smaller clutches (ANCOVA with binomial errors: between-final nest effect: $\chi^2_{57}=89.4$, $p=0.004$; fledging mass: $\chi^2_2=7.93$, $p<0.005$; natal clutch size: $\chi^2_1=10.1$, $p<0.002$). Open circles, 1995; filled circles, 1996. Number of chicks per group are given in the box at the top of the graph.

sample size than analysis in figure 1, since not all laying dates were known exactly). As chicks from different broods growing up in the same nest are compared, other effects of time of season on recruitment rate are unlikely. Territory size was measured in part of the population, and enlarged for some of the birds, but territory size of the biological parents also did not explain a significant part of the variation in recruitment rates within broods ($\chi^2_1=0.89$, $p=0.35$, effect tested with final brood size, natal clutch size and fledging mass). If anything, the effect was that chicks from larger natal territories had slightly lower recruitment chances.

If chicks originating from larger clutches recruit better because they are genetically better survivors, then one expects that their biological parents are also better survivors. If recruitment rate was analysed within the nest for dependence on the number of the biological parents that survive to the next breeding season (thus 0, 1, or 2), then

there is no evidence that recruitment rate was positively correlated with the number of surviving biological parents (ANCOVA with binomial errors: between-final nest effect: $\chi^2_{56}=87.4$, $p=0.005$; fledging mass: $\chi^2_1=9.35$, $p=0.002$; natal clutch size: $\chi^2_1=8.75$, $p=0.003$; number of surviving parents: $\chi^2_1=2.74$, $p=0.12$). If anything, the effect was that juveniles with more surviving parents were poorer at recruiting, so no resemblance existed between survival of parents and the recruitment of their genetic offspring.

4. DISCUSSION

The within-year variation in clutch size was not related to the parental capacity to raise offspring, as revealed by the experiment in which all the broods were manipulated to contain nine chicks. This is consistent with the data from the natural variation in clutch size. This experiment was based upon earlier experiments in which the same manipulation also did not yield evidence for individual optimization of clutch size (Both 1998). Thus, different studies on the same species do give evidence whether individuals do (Perrins & Moss 1975; Pettifor *et al.* 1988; Tinbergen & Daan 1990), or do not, optimize their clutch size according to their local circumstances (discussion on this subject will be published by J. M. Tinbergen and C. Both, see also Dhondt *et al.* (1990)).

Young from larger clutches recruited better than nest-mates from smaller clutches if raised under identical conditions. Norris (1993) showed, in a cross-fostering experiment, that great tit males with a more developed secondary sexual character had sons, but not daughters, with higher survival. In his experiment complete clutches were swapped between nests, thus the comparison was not between chicks that were raised under identical conditions. Both experiments suggest that there are either environmental effects before the chicks were exchanged, or that genetic differences exist in survival ability. We have no evidence of early maternal effects, because eggs were of similar size, as were the nestling mass at manipulation and the fledging mass. Genetic differences, on the other hand, seem to be unlikely for a trait that is so closely related to fitness (Fisher 1930; Gustafsson 1986). Genetic variation could not be shown, since chick recruitment was not related to the survival probability of their parents.

Chicks originating from larger broods recruit better if raised under the same conditions as chicks from a smaller original clutch size. If recruitment rate of chicks from unmanipulated nests is analysed there is no effect of the clutch size of their parents (data 1983–1990, $n=1568$, logistic regression: factor for year: $\chi^2_7=18.5$, $p=0.001$; fledging mass: $\chi^2_1=4.2$, $p=0.04$; clutch size: $\chi^2_1=2.9$; $p=0.09$; if anything, clutch size affected survival negatively). It thus seems that better recruitment is not an absolute value, but is related to the number and origin of nest-mates. This means that one of the basic life-history trade-offs, i.e. the trade-off between quality and quantity of offspring, cannot always be easily measured in the nest, because mass and size at fledging are not in all cases good predictors for viability. If parents are able to manipulate the competitive strength of their offspring in an adaptive way (Schwabl *et al.* 1997), then exchanging

chicks between different broods might not mimic the natural situation of an individual laying more or less eggs. Most of the classical work on clutch size optimization has not included the simultaneous optimization of number and quality of eggs, which will be one of the main challenges in future work on this subject.

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