The effect of variation in the laying interval on proximate determination of clutch size in the European Kestrel

José Miguel Aparicio


I propose that if laying interval varies depending on daily energy intake, then laying interval should also play a basic role in the determination of clutch size. A supplemental feeding experiment with European Kestrels Falco tinnunculus was performed to test this hypothesis. Supplemented females laid larger clutches with shorter intervals between eggs than control females, but did not take longer to complete their clutches. Clutch size was negatively correlated with mean laying interval. Clutch size also declined seasonally, but the effect of laying date disappeared when laying interval was controlled for statistically. Therefore, food supply affected clutch size through laying interval, not through laying date.


Numerous observational and experimental studies have found a positive relationship between a female’s nutritional state and clutch size (e.g. Jones and Ward 1976, Houston et al. 1983, Martin 1987, Arcese and Smith 1988, Nilsson 1991). This suggests that clutch size is constrained by feeding conditions. However, there is also evidence to the contrary (e.g. Arnold and Rohwer 1991). Thus, indeterminate layers are able to alter their clutch size in response to egg removal or addition (Cole 1917, and reviews in Klomp 1970, Kennedy 1991, Haywood 1993), by obtaining the protein and energy demands for their eggs from daily food intake (e.g. King 1973, Ojanen 1983, Meijer et al. 1989).

To understand the role of feeding conditions on the proximate control of clutch size it may be necessary to consider the factors which determine the cessation of egg-laying, which seems to be endocrinologically controlled. Cessation of laying is related to rising prolactin levels (e.g. Burke and Denison 1980, Opel and Proudam 1980, Lea et al. 1981). High levels of prolactin inhibit both the follicle-stimulating hormone (FSH) and the luteinizing hormone (LH), which are associated with follicle development and ovulation (Lehrman 1959, Camper and Burke 1977, Burke and Denison 1980, Zadworny et al. 1989). Prolactin levels increase gradually throughout the prelaying period and are further accelerated during the laying period (e.g. Lea et al. 1982, Meijer et al. 1990). In some species, this acceleration may be caused by tactile stimulation between the eggs and the brood patch(es) (e.g. El Halaoui et al. 1980, Hall and Goldsmith 1983, Hall 1987), but in other species (perhaps determinate layers) prolactin increases before egg-laying (Hector and Goldsmith 1985). In either case, females have a limited period of time in which to develop follicles before prolactin reaches a level at which LH and FSH are inhibited (see Meijer et al. 1990). I hypothesize that if follicle growth varies depending on nutritional state, then females in good condition should lay eggs at shorter intervals within the time available for egg-laying, than females in poor condition. Thus, feeding conditions would act as a proximate control on clutch size, but females would be able to respond to egg removal before prolactin levels start increasing rapidly.

Laying interval varies among species (e.g. Aslikeimer 1985) and within species (e.g. von Haartman 1990, Village 1990, Beissinger and Wallman 1991). Lack (1968) and Aslikeimer (1985) suggested that such variations may reflect underlying differences in the ability of birds to obtain energy for egg formation. Bryant (1975) found an increase in House Martin Delichon urbica laying intervals during periods of low food availability. Recently, Nilsson and Svensson (1993) showed that gaps in laying
Table 1. Laying interval, time taken by females to complete egg laying, and clutch size (mean ± sd) of food-supplemented and control Kestrels. Differences tested with Mann-Whitney U-test (two tailed probabilities).

<table>
<thead>
<tr>
<th></th>
<th>Supplemented</th>
<th>Control</th>
<th>U</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Laying interval (d)</td>
<td>2.04 ± 0.22</td>
<td>2.50 ± 0.65</td>
<td>33.5</td>
<td>0.02</td>
</tr>
<tr>
<td>Duration of laying (d)</td>
<td>8.4 ± 0.8</td>
<td>8.0 ± 1.2</td>
<td>56.5</td>
<td>0.36</td>
</tr>
<tr>
<td>Clutch size</td>
<td>5.1 ± 0.3</td>
<td>4.3 ± 0.7</td>
<td>36.0</td>
<td>0.04</td>
</tr>
</tbody>
</table>

(intervals between successive eggs longer than the usual of one day) were less frequent when Blue Tits Parus caeruleus females received supplementary food. Moreover, Watson et al. (1993) found that laying intervals decreased with clutch size in the Common Eider Somateria mollissima.

To test the effect of food availability on laying interval and clutch size, I performed a supplementary feeding experiment on a population of European Kestrels Falco tinnunculus. I compared mean laying interval of consecutive eggs, clutch size and the time to clutch completion between supplemented and non-supplemented birds. Moreover, I examined the relationships between laying date, laying interval and clutch size.

Methods

This study was conducted on a Kestrel population in the province of Cuenca, in Central Spain (40° 8' N, 2° 18' W). The area is an agricultural plain, that is cultivated mainly with cereals and sunflowers. In this area Kestrels breed in natural rock cavities and to a lesser extent in old corvid nests in trees. Their diet is chiefly composed of insects. However, during the courtship and incubation periods, females are regularly fed by their partners with lizards and small birds (J.M. Aparicio, unpubl.).

The supplementary feeding experiment was carried out during the spring of 1990. Breeding pairs were found several weeks before the onset of laying. Sixteen randomly selected pairs were supplemented for a minimum of 17 days before egg laying (44 ± (SD) 20 d). This period is nearly twice as long as the nine days that Kestrels require to develop their first egg (Meljer et al. 1989). Feeding ceased when the pair had completed laying. Kestrels were supplemented, every two days, with 120–150 g of whole chickens per pair. Yellow-coloured pellets indicated that the chickens were eaten by the Kestrels. The control group included 9 pairs; six other unsupplemented pairs had to be excluded because I was not able to record laying intervals for them.

Each nest was monitored every two days from the middle of April until the first egg was laid. After this, the nest was monitored daily until the clutch was completed. Visits to nests began two hours after sunrise. When females had not laid at the expected normal 2-d interval I revisited nests later in the evening, though no new eggs were found in the afternoon. Each egg was marked with a waterproof felt tip pen. I discovered no partial predation of clutches with marked eggs and it is unlikely that long intervals between eggs were recorded because unmarked eggs had been preyed upon during the short time (only a few hours) between egg-laying and nest-checking. Laying date is defined as the day the first egg was laid. I did not know exactly when the first egg was laid because nests were checked every two days before egg-laying. To estimate the time taken by each female to complete the clutch, I assumed that the first egg was laid at least two days before the second egg. This gives a 13% chance of an error of ±1 day in the estimation of time taken to clutch completion, i.e. half the probability (26%, see below) of a laying interval longer or shorter than two days. Clutch size was determined when laying had ceased for five consecutive days.

Results

Variation in laying interval between consecutive eggs

Kestrels laid their eggs at intervals varying between 1 and 4 days. The mean laying interval was 2.12 days (sd=0.54, n=66). The modal interval was 2 days (74.2%) and laying intervals of 1, 3 and 4 days occurred with frequencies of 7.6%, 16.7% and 1.5%, respectively. The mean laying interval varied from 1.67 to 4 days between pairs. Twelve of 25 pairs (48%) laid their eggs with mean intervals of 2 days.
Mean laying interval was significantly longer for control pairs than for food-supplemented pairs (Table 1). Therefore, laying interval seemed to be affected by daily energy intake during the egg-formation period.

Effect of laying interval and laying date on clutch size

The mean laying interval between consecutive eggs was negatively correlated with clutch size in supplemented nests \((r = -0.63; n = 16; P < 0.01)\), non-supplemented nests \((r = -0.79; n = 9; P < 0.01)\) and all nests combined \((r = -0.79; n = 25; P < 0.0001); Fig. 1\).

As expected, clutch size was positively correlated with the time taken to complete the clutch \((r = 0.69; n = 25; P = 0.0001)\); however, this does not prove that clutch size is determined by the time taken to complete the clutch rather than vice versa. To test these two alternatives, I compared the time taken to complete the clutch between supplemented and control pairs. Supplementary food caused larger clutch sizes (Table 1), so I predicted that if clutch size determines duration of laying, then supplemented females should take more time than control pairs to complete their clutches. However, there was no significant differences in duration of laying (U-test, two-tailed: \(U = 56.5; P = 0.36\); Table 1).

Prolactin levels in Kestrels may increase gradually in spring (Meijer et al. 1990). Thus, the time available for egg-laying may decrease as laying date advances. Therefore, laying date could affect clutch size. In this study there was a weak negative trend between laying date and duration of laying \((r = -0.33; n = 25; P = 0.1); Fig. 2\). Hence, I should expect a weak influence of laying date on clutch size. In fact, the negative relationship of clutch size to laying date \((r = -0.42; n = 25; P = 0.036); Fig. 3\) may be because food supply influenced both laying date and clutch size (Aparicio in press), because the effect of laying date on clutch size disappears when laying interval is also considered in a multiple regression (Table 2).

**Discussion**

**Proximate determination of clutch size**

I hypothesized that if laying interval varies in relation to feeding conditions, then food supply should play a basic role in the determination of clutch size through laying interval, because females in a better nutritional state can lay larger clutches in the time available for egg-laying. The results of this study support this hypothesis. Females receiving supplementary food laid more eggs at shorter intervals, than did control females, yet there was no difference in the duration of laying between treatments. This result indicates that the duration of laying may affect clutch size, but not vice versa. The direction of this causal relationship may result from a necessary proximate control of clutch size. Furthermore, there may be selective pressures that determine this relationship. The viability of unincubated eggs declines with time so clutch size might be constrained by the amount of time available for egg laying (Arnold et al. 1987, Veiga 1992, Arnold 1993, Veiga and Viñuela 1993).

King's (1973) model of energy allocation to egg formation could explain why differences in feeding conditions lead to variations in laying interval. The model shows that in such birds where a succession of follicles begin development at intervals of \(n\) days, the daily energy demand for the formation of the clutch reaches a peak that is independent of clutch size. This peak is reached \(n\) days before laying date and is sustained for as long as new follicles grow. If a female cannot take in enough

<table>
<thead>
<tr>
<th>Independent variable</th>
<th>(b)</th>
<th>(t)</th>
<th>(P)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Laying date</td>
<td>-0.015</td>
<td>1.24</td>
<td>0.227</td>
</tr>
<tr>
<td>Laying interval</td>
<td>-1.147</td>
<td>5.29</td>
<td>&lt;0.0001</td>
</tr>
</tbody>
</table>

Model: \(F_{2.22} = 20.5; P < 0.0001\).
Table 3. The modal laying interval and the effects of supplementary food on clutch size.

<table>
<thead>
<tr>
<th>Species</th>
<th>Effect of sup. food on clutch size</th>
<th>Laying interval (days)</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>Falco amandae</td>
<td>- ns</td>
<td>1</td>
<td>Hill (1988)</td>
</tr>
<tr>
<td>Falco amandae</td>
<td>1.0 s</td>
<td>1</td>
<td>Arnold (1994)</td>
</tr>
<tr>
<td>Larus cullio</td>
<td>0.9 s</td>
<td>1</td>
<td>Carlson (1989)</td>
</tr>
<tr>
<td>Prunella modularis</td>
<td>0.2 s</td>
<td>1</td>
<td>Davies and Lundberg (1985)</td>
</tr>
<tr>
<td>Ficedula hypoleuca</td>
<td>- ns</td>
<td>1</td>
<td>J.J. Sanz and M. Moreno (unpubl.)</td>
</tr>
<tr>
<td>Parus major</td>
<td>- ns</td>
<td>1</td>
<td>Källander (1974)</td>
</tr>
<tr>
<td>Parus major</td>
<td>-1.0 ns</td>
<td>1</td>
<td>Clamens and Isenman (1989)</td>
</tr>
<tr>
<td>Parus caeruleus</td>
<td>0.6 ns</td>
<td>1</td>
<td>Clamens and Isenman (1989)</td>
</tr>
<tr>
<td>Parus caeruleus</td>
<td>0.0 ns</td>
<td>1</td>
<td>Nilsson and Svensson (1993)</td>
</tr>
<tr>
<td>Parus palustris</td>
<td>0.9 s (1)</td>
<td>1</td>
<td>Nilsson (1991)</td>
</tr>
<tr>
<td>Parus cristatus</td>
<td>0.3 ns</td>
<td>1</td>
<td>von Brömssen and Jansson (1980)</td>
</tr>
<tr>
<td>Parus montanus</td>
<td>0.3 ns</td>
<td>1</td>
<td>von Brömssen and Jansson (1980)</td>
</tr>
<tr>
<td>Melospiza melodia</td>
<td>0.1 s</td>
<td>1</td>
<td>Smith et al. (1980)</td>
</tr>
<tr>
<td>Melospiza melodia</td>
<td>0.5 s</td>
<td>1</td>
<td>Arcese and Smith (1988)</td>
</tr>
<tr>
<td>Agelaius phoenicus</td>
<td>0.0 ns</td>
<td>1</td>
<td>Ewald and Rothen (1982)</td>
</tr>
<tr>
<td>Agelaius phoenicus</td>
<td>- ns</td>
<td>1</td>
<td>Winberger (1988)</td>
</tr>
<tr>
<td>Xanthopis xanthocephalus</td>
<td>0.1 ns</td>
<td>1</td>
<td>Källander and Karlsson (1993)</td>
</tr>
<tr>
<td>Streptopelia virgata</td>
<td>0.1 ns</td>
<td>1</td>
<td>Högestedt (1981)</td>
</tr>
<tr>
<td>Pica pica</td>
<td>0.5 s (1)</td>
<td>1</td>
<td>Hochachka and Boag (1987)</td>
</tr>
<tr>
<td>Pica pica</td>
<td>0.4 s</td>
<td>1</td>
<td>Knight (1988)</td>
</tr>
<tr>
<td>Pica pica</td>
<td>- ns</td>
<td>1</td>
<td>Dhindsa and Boag (1990)</td>
</tr>
<tr>
<td>Pica pica</td>
<td>0.7 s</td>
<td>1</td>
<td>Yom-Tov (1974)</td>
</tr>
<tr>
<td>Accipiter nisus</td>
<td>0.8 s</td>
<td>2</td>
<td>Newton and Marquiss (1981)</td>
</tr>
<tr>
<td>Falco tinunculus</td>
<td>0.8 s (1)</td>
<td>2</td>
<td>Dijkstra et al. (1982)</td>
</tr>
<tr>
<td>Falco tinunculus</td>
<td>- s (2)</td>
<td>2</td>
<td>Meijer et al. (1988)</td>
</tr>
<tr>
<td>Falco tinunculus</td>
<td>0.8 s</td>
<td>2</td>
<td>This study</td>
</tr>
<tr>
<td>Aegolius funereus</td>
<td>0.9 s</td>
<td>2</td>
<td>Körpinäk (1989)</td>
</tr>
<tr>
<td>Aegolius funereus</td>
<td>0.9 s</td>
<td>2</td>
<td>Hörnfelt and Eklund (1990)</td>
</tr>
<tr>
<td>Lanius fuscus</td>
<td>0.2 s (1)</td>
<td>2</td>
<td>Bolton et al. (1992)</td>
</tr>
</tbody>
</table>

ns: non-significant effect; s: significant effect. (1) Significant differences in one of two years. (2) Significant differences in late feeding experiment.

Nutrients during the peak investment period for egg formation, she might reduce this peak by prolonging the time of formation of each egg, thereby increasing the laying interval between consecutive eggs.

Laying interval in Kestrels frequently deviated from the modal interval of two days. These variations in laying interval accounted for 63% of the variation in clutch size. Laying interval may be less likely to affect clutch size in other species which usually lay one egg each day. In these species, such effects will only occur when low intake prolongs the interval between eggs (e.g. Bryant 1975, von Haartman 1990). However, laying interval cannot be shortened when feeding conditions are better. Thus, variation in laying interval should not be as frequent as in the Kestrel (see Nilsson and Svensson 1993). The determination of clutch size could be the same in both cases. However, the effect of food supply should be smaller if variations in laying interval are so too. This would explain why only 6 of 23 supplementary feeding experiments found significant effects on clutch size in species which lay one egg per day (Table 3). In such species, it is probably easier to find an effect by restricting the food available to the birds or their capacity to forage (e.g. Slagsvold and Lifjeld 1988, 1990). By contrast, when laying interval was longer and probably more variable, every experimental study showed a significant effect of supplemental feeding on clutch size (Table 3).

Daan et al. (1988) suggested that laying date may have a proximate effect on clutch size. In the population studied here, clutch size declined with the progress of the season. However, the effect of laying date on clutch size disappeared when laying interval was controlled for statistically. Therefore, any relationship between clutch size and laying date in this population seems to be determined by the effect of food supply on both, by advancing laying date and enlarging clutches, rather than by the effect of laying date on clutch size. In fact, the typical seasonal decline in clutch size was not observed for supplemented pairs (Aparicio in press). However, other supplemental feeding experiments have shown that laying date has an effect on clutch size that is independent of food availability (e.g. Meijer et al. 1988, Hörnfelt and Eklund 1990, Nilsson 1991, Arnold 1994). However, apart from Arnold (1994) who did not find significant differences in laying rates, and Nilsson and Svensson (1993), no other study has investigated the effect of supplementary feeding on laying interval. There remains ample scope for such studies in other species.
Linking factor

Fig. 4. The adjustment between proximate and ultimate determination of clutch size. The continuous line represents optimal clutch size and broken lines represent two cases of feasible clutch size in relation to a linking factor (e.g., the rate of energy intake during egg formation). In case A, the feasible clutch size is larger than the optimal clutch size, and natural selection acts advancing the time at which the prolactin levels are accelerated and/or shortening the time taken to complete the clutch and/or increasing the amount of energy per egg, whilst in the case B natural selection acts in the opposite way.

Compatibility of proximate and ultimate determination of clutch size

At least in nidicolous birds, the proximate mechanism of clutch size determination must adjust clutch size to the number of nestlings that can be adequately fed, since clutch size is individually optimized in relation to the parental capacity to rear young (e.g., Höglund 1980, Gustafsson and Sutherland 1988, Pettifor et al. 1988, Lindén 1990, Pettifor 1993). Such an adjustment requires a link between the proximate and ultimate factors that control clutch size. This link must combine two functions: to be a cue for the optimal brood size, and to constrain feasible clutch size. The rate of energy intake during the prelaying and laying periods may comply with both requirements, because it affects laying interval, and may be a realizable cue when future food supply depends on previous conditions (e.g., Benkman 1990, Donázar et al. 1992, Tye 1992).

The linking factor (rate of energy intake) would be related to optimal and feasible clutch size. However, to achieve the individual adjustment of clutch size, natural selection must have operated so as to achieve minimum differences between both relationships (Fig. 4). Natural selection could act on any factor involved in the mechanism of proximate determination, operating on the time at which the prolactin levels are accelerated, on the acceleration value of prolacitin levels, or on the invested amount of energy per egg. The two first would have an effect on time available for egg-laying, and the last on laying interval. Thus, clutch size may be constrained by daily energy intake through laying interval, but such constraints may also allow individual optimization of clutch size.

Acknowledgements – I thank T.W. Arnold, G.R. Bortolotti, D.C. Houston, R. Johnston, J. Moreno, and R.A. Pettifor for helpful comments on this manuscript. I was supported by a grant from the Ministerio Español de Educación y Ciencia.

References

Aparicio, J. M. (in press). The seasonal decline in clutch size: an experiment with supplementary food in the Kestrel Falco tinunculus. – Oikos.


Jones, P. J. and Ward, P. 1976. The level of reserve protein as the proximate factor controlling the timing of breeding and clutch-size in the Red-billed Quelea, Quelea quelea. – Ibis 118: 547–574.

(Received 26 November 1993, revised 1 April 1994, accepted 19 May 1994.)