The energetic costs of egg heating constrain incubation attendance but do not determine daily energy expenditure in the pectoral sandpiper

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Heating eggs during incubation may be relatively energetically costly, affecting the outcome or number of breeding attempts. We determined the effect of reduced egg heating costs on nest attendance, change in body mass, and daily energy expenditure (DEE using the doubly labeled water technique) by heating nests of pectoral sandpipers. We also considered ground temperature, which may influence overall incubation costs, and mass reserves and stage of incubation, which may influence an individual’s ability to respond to changes in overall incubation cost. The total proportion of time spent in attending the eggs was significantly greater in nests that were experimentally heated (3.6% or 52 min daily), and this effect was significantly greater at low ground temperatures (14.7% or 211.7 min daily). Mass change was independent of experimental heating when controlling for attendance, although mass loss rate was greater for birds that attended more (for every 10% increase in daily proportion of attendance 0.12 extra grams of body mass were lost per hour), and overall daily attendance increased by 0.5% for every extra 1 g of body mass. DEE was greater for birds that had the higher rates of mass gain (for every 0.1 g of mass gained per hour, DEE increased by 20.5 kJ per day) but was independent of experimental heating when controlling for attendance. Overall, the results suggest that females are constrained from attending more by their energy reserve levels being depleted at least partly by the costs of egg heating, but these costs probably do not determine DEE, as costs off the nest may far exceed those incurred while sitting.

Breeding in the arctic is clearly energetically demanding: pectoral sandpipers had an average DEE of 361.1 ± 8.9 kJ per day, equivalent to 6.1 times basal metabolic rate (n = 24 birds). Key words: daily energy expenditure, incubation, nest attendance, pectoral sandpiper. [Behav Ecol 15:498–507 (2004)]

Incubation costs may be especially high in species nesting in cold environments, where heat is rapidly lost from the eggs (Tinbergen and Williams, 2002; Williams, 1996), and in species in which only one parent incubates (uniparental incubators). In this case, the energetic bottleneck caused by the cost of heating the eggs may be exacerbated because the energetic intake of the incubating parent may be constrained by the need to remain on the nest (Drent et al., 1985). Finally, uniparental incubators that must leave the nest to feed are likely to experience a further cost, caused by the need to rewarm the cool clutch upon return to the nest (Williams, 1996). In the present study, we determine the effect of reduced egg heating costs on the time and energy budgets of a uniparental shorebird incubating in extreme arctic conditions, a system for which we predict egg heating costs are likely to be at a very high level relative to those of most other bird species (Tinbergen and Williams, 2002).

If, in uniparental incubators, maintaining clutch temperature is a limiting component of a breeder’s energy budget, then changes in the cost of egg heating should result in changes in some other major component of the breeder’s resource allocation. A reduction in incubation costs could result in increased nest attendance (Bryan and Bryant, 1999), which will confer fitness benefits because the duration of the

As individuals have finite resources at their disposal, the energetic costs of each reproductive stage may affect the outcome of the current breeding attempt or the number or outcome of future attempts (Stearns, 1992). Strategies that minimize the ratio of costs to benefits should be favored at all life-history stages, and thus, an understanding of the energetic demand of each reproductive activity is necessary to understand overall patterns of reproductive behavior and the resultant life histories. Birds have proved valuable models for dissecting patterns of resource allocation, both within and between reproductive phases (Monaghan and Nager, 1997; Stearns, 1992). Recent studies suggest that incubation (the process by which birds regulate egg temperature) represents an important energetic cost that may constrain current and future reproductive performance (Monaghan and Nager, 1997; Perrins, 1970; Reid et al., 2002b).
incubation period may depend upon nest attendance (Spiers and Baummer, 1990) and longer incubation periods may reduce breeding success (see Tombre and Erikstad, 1996). Alternatively, incubating birds may increase their body reserves if this provides greater fitness benefits than does increasing nest attendance (see Reid et al., 2002b; Slagsvold and Johansen, 1998). Variation in a significant energetic cost of incubation could also be expressed as variation in daily energy expenditure (DEE), but only if birds do not compensate for variation in the cost of incubation by varying their allocation to other activities (Tinbergen and Williams, 2002). In this study we test whether the cost of egg heating during the incubation stage is sufficient to constrain time or energy allocated to incubation by testing whether nest attendance increases, body reserves increase, or DEE is decreased when the cost of egg heating is experimentally decreased. The present study is one of the first to experimentally manipulate an energetic cost of incubation while measuring DEE in a natural system, in which energetic costs are likely to be high.

We experimentally reduced the costs of egg heating by experimentally warming pectoral sandpiper nests (Calidris melanotos), an arctic breeding shorebird in which only the female incubates. We determined the effect of the reduction in egg heating cost on nest attendance, change in mass, and DEE (as revealed by doubly labeled water [DLW] analysis). We considered the effects of ground temperature because this is likely to influence overall incubation costs (Williams, 1996). We also considered mass reserves (see Aldrich and Raveling, 1983) and stage of incubation (Skutch, 1962), which may influence an individual’s ability to respond to changes in egg heating cost. We predict that if the cost of heating eggs significantly affects a female’s ability to allocate resources to incubation, then reducing egg heating costs will increase attendance, reduce mass loss for any given attendance period, reduce DEE, or any combination of the three. However, exact predictions of which variables will change and their direction of change are difficult to make because changes in each of the three variables will affect the other two, and so, compensation is possible. For example, reducing egg heating costs may reduce incubation costs significantly but may not change attendance because a bird may choose to reduce its rate of mass loss while incubating and, thus, its overall DEE (because less mass needs to be regained when feeding off the nest). Similarly, if one activity (e.g., time off the nest) is replaced with one that is equally energetically demanding (such as incubation), then we would expect to see no change in DEE. We can, however, conclude that if none of the three variables are affected by reducing egg heating cost, then this cost is relatively unlikely to be a major constraint.

METHODS

We studied pectoral sandpipers breeding at Barrow, Alaska (71°10’ N, 156°42’ E), from 10 June–4 July 2000. The study area has been described in detail elsewhere (Norton, 1972). We found 36 nests, a sample of which were used in this study. We experimentally heated 11 nests to reduce the amount of energy needed to heat the eggs by a sitting female and successfully measured how nest heating affected nest attendance, change in mass, and DEE compared with values for females incubating on 17 unmanipulated control nests. We also measured a number of weather variables, but particularly ground temperature that may have affected the relative importance of the reduced egg heating costs. Finally, we measured two variables that may have affected an individual’s ability to respond to reduced egg heating costs: stage of incubation and initial body mass. All procedures were carried out under relevant US federal and state permits.

Pectoral sandpipers are uniparental incubators in which the female only incubates for about 85% of the time, depending on weather conditions (Norton, 1972). The clutch size is typically four, the incubation period is 21–23 days, and nests are on slightly raised sites in wet sedge tundra and gradually become covered by growing vegetation (Holmes and Pitelka, 1998).

Experimental treatment

To investigate the consequences of a reduction in costs of egg heating, a nest was experimentally heated with a nonmanipulated nest acting as a control. Pairs of nests were randomly assigned to each group with the intention of having an experimental and a control nest monitored at the same time in a matched-pair format so that environmental conditions would be the same when the nests were compared. The experiment was carried out over 11 days from 21 June–2 July 2000, with a mean of 2.4 nests (range = 1–5) being inspected per day. Each experimental nest was heated for 48 h (the experimental period) with attendance, mass change, and DEE being estimated for the second 24-h period of heating (the sampling period).

Nests were experimentally heated by placing chemical hand warming pads under the nest cup. The pad was insulated with foam and aluminium foil on its lower side to reduce the rate of conductive heat loss to the ground. The eggs were removed from a nest, and the warming pad was inserted into a slit, cut below the nest lining. The vegetation was pushed back into place and the eggs replaced. The procedure took 3–5 min and left the nest in a state similar to its original condition. When the sandpiper was captured for measurements at the start of the sampling period (day 2), the heating pad was replaced with a new one. Control nests were not manipulated in any way. This means that there were two potential experimental effects in operation: (1) the effect of interest, nest heating, and (2) an effect of disturbance caused by the addition of heating pads. Control nests were not manipulated (e.g., with the addition of dummy nonheating pads) to minimize the disturbance to the birds. At the start of the experiment, we had no idea whether any manipulation would lead to desertion or nest failure owing to predation, and we did not wish to risk losing all of the study nests if this was the case. This means, however, that the effects of increased disturbance that may have reduced attendance, and may have reduced the effect of nest heating that was expected to increase nest attendance. The importance of this potential confounding effect is considered in the first section of the Results.

The effect of the heating pad on egg temperature was determined by five experiments on five different pairs of artificial pectoral sandpiper nests constructed to mimic natural nests (the validity of this is discussed in Reid et al., 2002a). A single pectoral sandpiper egg taken from a deserted clutch was placed in both nests: the egg was blown and filled with plaster of Paris with a fine wire thermistor inserted into the center. Pectoral sandpipers have a clutch of four eggs, and therefore, our estimates of the effects of experimental nest heating on a single egg do not take into account the effects of the larger volume and the differing surface area-to-volume ratio (which will vary according to how the four eggs pack together) in a normal clutch. Norton (1973) found that the absolute rate of heat loss for wader species at Barrow was greater for three egg than for four egg clutches, suggesting that our single egg measure of the effects of nest heating may be an underestimate as larger clutches will lose heat at a slower rate. The thermistors were connected to Tinytag data loggers.
(Gemini Dataloggers Ltd.) so that the eggs had a length of insulated fine wire cable protruding from each egg, connecting them to the data logger. Egg temperature was recorded every 5 s over a 24-h period. To attempt to simulate the route of heat loss from the eggs during steady-state incubation, the artificial nests were covered with a layer of expanded polystyrene, topped with aluminium foil, to try to ensure that heat was exchanged only with the surrounding ground. If the sitting bird’s body does not completely cover the nest, heat may also be lost to the air outside the nest (see Walberg and King, 1978). However, this is unlikely to be a major route of heat loss from pectoral sandpiper nests, which are sited in dense graminoid tussocks (Holmes and Pitelka, 1998), which will reduce wind speed and forced convective cooling. In one nest (this was swapped for each experiment) a heating pad was inserted underneath the cup exactly as with the natural nests described above. The difference between the temperatures of the eggs in the unheated and heated nests was calculated hourly over a 24-h period. There was a clear effect of heating (Figure 1): egg temperatures in the heated nests were raised by several degrees throughout the 24-h period, although heating was most pronounced in the first 12 h. Overall, there was an experimental increase in mean egg temperature of 11.4°C (95% confidence limit [CL] = 9.8–13.1, n = 3 pairs of nests) over the 24-h period. If we assume that the rate of heat loss from the clutch is simply proportional to the difference between the target egg temperature for incubation (i.e., 36°C) and the equilibrium temperature of the unincubated egg (not exposed to solar radiation), then the energetic costs of heating the eggs during steady-state incubation may have been reduced by 36.8% (95%CL = 30.5–45.2). However, the energetic costs of intermittent incubation also consist of egg reheating costs; although these costs were also likely to be reduced by the experimental nest heating, we cannot determine the exact reduction in overall energetic costs of incubation of the experimental treatment.

**Attendance**

Nest attendance was measured by using a flexible-tipped temperature probe connected to a TinyTalk datalogger. The probe was placed in the center of the clutch, and nest temperature was recorded every 30 s. As nest temperatures were above 30°C while the female was incubating the clutch but air temperature rarely exceeded 10°C, nest temperature changed rapidly when a bird departed from or returned to the nest; thus, the temperature traces provided a clear record of attendance that was accurate to the nearest minute. In a previous study on arctic-alpine nesting dotterel (*Charadrius morinellus*), using an identical set-up, we achieved 100% accuracy for 58 opportunistic ground truthing records when the bird was off and 98.9% for 91 records when attended (Holt S, unpublished data; see also Reid et al., 1999). The times that the female departed from and returned to each nest were then used to calculate the duration of each incubation bout and each trip off the nest; the sum of the total time spent incubating/the total sampling period was used to determine mean nest attendance. The total experimental sampling period for the 28 nests was 24.4 ± 0.3 h (range = 22.7–28.9 h): any biases in attendance sampling owing to diurnal variation in attendance were randomly distributed across the experimental and control group (sampling periods of heated and control nests, \( b_0 = -0.2, p = .76 \)).

Nest attendance was measured for a minimum of 24 h at 46 nests before any experimental manipulation, including the experimentally manipulated (\( n = 11 \)) and concurrent control nests (\( n = 17 \)) throughout the experimental period.

**Change in mass**

Incubating females were captured by using walk-in nest traps and were ringed with a metal ring covered with colored fluorescent tape, allowing individuals to be recognized by sight over the duration of the study. Females were weighed to the nearest 0.5 g by using a Pesola spring balance at the start and at the end (24.4 ± 0.3 h later) of the sampling period. Twenty-one individuals were caught and ringed several days before the experiment and were additionally weighed at this time.

**Daily energy expenditure**

DEE was estimated for 28 female pectoral sandpipers by using the single-sample DLW technique (Webster and Weathers, 1989; for theory and calculations, see Speakman, 1997). At the start of the 24-h sampling period, we injected the bird with 0.30–0.39 ml of DLW (scaled to the bird’s mass) consisting of 33.3% by volume of 99% enriched \( ^{2}H_2O \) (deuterium oxide) and 66.6% by volume of 95% enriched \( ^{18}O \) (measured by volume). The bird was injected subcutaneously in the ventral region and was immediately released. From capture to release took on average less than 3 min. Most females returned to the nest within a few min (23.3 ± 6.5 min after injection: there was no significant difference in return times between experimental and control birds, \( b_0 = 0.7, p = .48 \)). The 28 females sampled were then recaptured 22.9–29.5 h later. We took 5 × 5 μl of blood from the brachial vein in glass capillaries, sealed immediately using a blowtorch. The bird was kept in the hand until all bleeding had stopped (usually 1–2 min) and then released. Initial equilibrium concentrations of the injected DLW were estimated from a further six pectoral sandpipers. These birds were caught and injected as above, and then held in the dark in a cardboard box for 60 min after the injection to allow the DLW to equilibrate with the bird’s body water. The bird was then blood-sampled and released. No further data were taken from these six birds other than monitoring their subsequent nest attendance to determine the effects of blood sampling on incubation behavior.
We used the single sample DLW technique, in which the average initial equilibrium concentration of the labeled water is estimated from individuals that are not subsequently included in the experiment rather than from the experimental individuals themselves, to minimize disturbance to the experimental birds (see Williams and Vézina, 2001). This proved to be a valid decision because at the end of the study we found that the blood sampling required to estimate the initial concentration significantly altered attendance behavior. Mean (±1 SE) trip length for a bird directly before (sampled for 23.3 ± 0.6 h) and after (sampled for 14.3 ± 2.1 h) blood sampling was 8.6 ± 1.2 min and 35.5 ± 10.8 min, respectively, a mean increase of 26.9 min (matched-pairs t test comparing n = 23 nest means, t22 = −2.4, p = .02). This increase in mean trip length after blood sampling also occurs when only nonexperimental nests were compared, so removing any potential confounding effects of the end of the experimental heating period (an increase of 42.6 ± 16.9 min, matched-pairs t test comparing n = 14 nest means, t13 = −2.3, p = .025). There was, however, no increase for birds that had been injected but not been bled; the mean trip length for a nonbled bird on two consecutive days was 7.3 ± 0.6 min on the first day and 7.4 ± 1.2 min on the second (matched-pairs t test, n = 41 nests sampled for attendance at least 2 days before blood sampling if at all, t40 = −0.1, p = .91). The mean difference between the changes in mean trip length in the two groups was an increase in 26.9 ± 10.8 min for birds that had been bled (t test, n = 23 birds that were bled compared with n = 9 different birds that only had their natural nest attendance measured and that were never experimental birds, t22.0 = 2.4, p = .02).

Blood samples were analyzed 4 months later in Natural Sciences and Engineering Research Council (NERC) Scientific Services Life Sciences Community Stable Isotope Facility. Oxygen was liberated from water in plasma and converted to CO₂ (a form suitable for mass spectrometric measurement of ¹⁸O/¹⁰O) using the guanidium hydrochloride technique (Tatner and Bryant, 1988). Triplelicate analyses of all enriched ¹⁸O standards used to calibrate the field sample enrichment showed repeatability (Lessells and Boag, 1987) of these measurements to be extremely high (r = .9995). Moreover, there was no significant difference between mean measured parts per million (ppm) and the independently calibrated value (matched-pairs t test k0 = 0.52, p = .96). Deuterium/hydrogen (D/H) ratio was measured by vacuum distillation of water from the blood sample, followed by reduction to hydrogen gas (a form suitable for mass spectrometric analysis) over chromium (Donnelly et al., 2001).

Background concentrations were 0.1988 atom percent for H₂¹⁸O and 0.0145 percent for H₂O. These values were the lowest of the values (0.19884 and 0.20089 H₂¹⁸O and 0.0145 and 0.0146 for ²H₂O) obtained from two blood samples from birds that had unusually low but similar value (i.e., clear outliers), suggesting that the values obtained reflected background levels. For these two samples, the isotope ratios were assumed to have decayed to background before we resampled the birds. These values were checked by plotting our empirical final values of both H₂¹⁸O and ²H₂O against time since injection and determining the exponential decay function of best fit (see Bearhop et al., 2002). The predicted values for the background levels were 0.19884 for H₂¹⁸O and 0.01450 for ²H₂O. Background values were further checked by comparison with all available data on H₂¹⁸O and ²H₂O concentrations in rainwater for Barrow (from http://isolis.iaea.org) from June–July, 1964–1966. These were 0.1974 ± 0.00018 and 0.0139 ± 0.00083, respectively. If these meteoric values are corrected for expected concentrations in avian blood (see Tatner, 1990), then blood concentrations would be expected to be approximately 0.1986 for H₂¹⁸O and 0.0135 for ²H₂O. Finally, we checked our chosen background values against empirical values reported for two temperate terrestrial bird species of similar size that feed on freshwater invertebrates, the dipper, Cinclus cinclus (0.1991 for H₂¹⁸O and 0.0146 for ²H₂O) and the common sandpiper, Actitis hypoleucos (0.1995 for H₂¹⁸O and 0.0149 for ²H₂O; Tatner, 1990). The background values chosen make no difference to the experimental analyses presented here because we are interested in relative change between two experimental groups in which the same background level applies to both groups. Calculations of absolute DEE, however, depend crucially on the choice of background values, and so where absolute values were calculated, we used these alternative background values to perform a sensitivity analysis to determine the effects of using one particular background value rather than another.

The percentage body water calculated from estimates of the dilution space of the sample of six birds blood sampled after 1 h was 76.2 ± 0.02% (n = 6); the six birds had a mean enriched isotopic concentrations of 0.6715 ± 0.0097 for H₂¹⁸O and 0.2958 ± 0.0061 for ²H₂O. We estimated DEE by using a respiratory quotient value of 0.72 and an energy equivalent of 27.1 kJ/L CO₂ (Piersma and Morrison, 1994). We estimated DEE by using the following standard equations:

\[
\text{Isotope turnover rate } k^{18} = \text{ ppm h}^{-1} = \ln(\text{initial isotope excess}) - \ln(\text{final isotope excess})/\text{sample interval}
\]

Size of body water pool \(N\) (mMol)

\[
N = \text{body mass} \times \text{body water fraction} \times (1000/18)
\]

\[
rCO_2 (\text{mMol h}^{-1}) = (N/2.08)(k^{18} - k_0) - (0.015 \text{ kDa } N)
\]

ADMR (em³ CO₂ g⁻¹ h⁻¹) = (rCO₂ × 22.4)/body mass

DEE (kJ d⁻¹) = ADMR × body mass × 24 × 27.1/1000

We converted DEE to an estimate of a multiple of basal metabolic rate by using the equations developed by Kersten and Pietsma (1986), where

\[
\text{BMR (Watts)} = 5.06 \times \text{ (body weight in kg)}^{0.729}
\]

Variables that may have affected the experiment

Three major kinds of variables that may have affected the energetic costs of an incubating bird, and therefore the results of the experiment, were considered: weather variables, incubation stage, and mass reserves.

Hourly measures of the weather variables air temperature, wind speed, global solar radiation, and rainfall were obtained from the Point Barrow Observatory for the National Oceanic and Atmospheric Administration, which is situated within the study area, and daily mean values were calculated. Ground temperature was measured each minute by using a temperature probe positioned 5 cm below the ground (the approximate level of a pectoral sandpiper egg) attached to a Tinytag data logger, and daily mean values were calculated. As there was only 1 day of rain during the study and very little total rainfall, effects of rainfall were not further considered. All weather variables were correlated; for example, air temperature was positively correlated with ground temperature (\(r = .91, p < .001\)), global solar radiation (\(r = .16, p < .001\)), and wind speed (\(r = .16, p < .001\)); all tests n = 541–576 available hourly means over the 24-day study period. Ground temperature was considered a priori to be the most important variable to influence the experimental effect of nest heating because egg heating is likely to be more demanding when the
Eggs were weighed to the nearest 0.1 g by using a portable electronic balance, and their maximum length and breadth was measured to 0.1 mm by using dial calipers. The egg density index was calculated as [mass/(breadth\(^2\) x length)].

The regression relationship between the mean egg density index for a clutch and the number of days from first egg date to the day of weighing was: 
\[ y = -0.0000033x + 0.00050, \quad F_{1.15} = 152.4, \quad R^2 = .90. \]

Eggs were weighed to the nearest 0.1 g by using a portable electronic balance, and their maximum length and breadth was measured to 0.1 mm by using dial calipers. The egg density index was calculated as [mass/(breadth\(^2\) x length)].

Experimental nests were sampled on average 12.1 days from first egg date to the day of weighing. Nests were discovered with less than four eggs, and so their date of completion was known. By weighing and measuring the eggs in these nests at intervals during the incubation period, we derived a relationship between mean egg density for a clutch and the time since the day the first egg was laid (Figure 2). We used this relationship to estimate the first egg date of nests that were only discovered after laying had been completed. Experimental nests were sampled on average 12.1 ± 0.8 days after the first egg was laid (range = 4–20 days): the period between first egg date and the time of an experiment on a nest was termed “day of incubation” and was put into all models to statistically control for any changes in attendance because nests were sampled at different times with respect to their first egg date.

Body reserves were estimated by measures of mass. There were two major confounding variables of mass as a measure of body reserves: variation in body size and changes in reserves as a function of incubation stage rather than the experiment. On capture, we measured the maximum wing chord of the bird (according to method of Prater et al., 1977) and then controlled for the effects of body size on reserve levels by dividing mass by the cube of wing length (see Piersma, 1984). A female’s body mass also declined with time since the first egg was laid (Figure 3).

RESULTS

Across the 46 nests for which attendance was monitored before conditions were experimentally altered, mean attendance was 81.5 ± 0.6%. On average, females made 1.2 ± 0.06 trips from the nest per hour, with median trip lengths of 6 min (range = 1–236 min) and incubation bout lengths of 18 min (range = 1–925). There was diurnal variation in attendance with few trips off the nest between 2000 and 0400h (median = 2 trips, range = 1–11), more trips between 0400 and 1200 h (median = 9, range = 1–23), and greatest trip frequency between 1200 and 2000 h (median = 17, range = 6–25).
The effects of the disturbance due to insertion of heating pads under the experimental nests was investigated by comparing the proportion of attendance for the 2-h period immediately after insertion of the first heating pad with that of exactly the same 2-h period 24 h previously when the nest was undisturbed. If the insertion of a heating pad affected attendance because of the disturbance that placing it entailed, we would predict that the proportion of attendance would be lower immediately after insertion compared with an un-disturbed period (at the same time of day). This was not the case, however, and there was no evidence for significant effects on attendance caused by the placement of the nesting pads alone: the mean difference was 1.1 ± 3.4 min between the period just after pad insertion and the control period 24 h before (t_{11} = -0.3, p = .75). The time that a bird was kept off the nest during pad insertion was relatively small (9.9 ± 2.9 min from first arrival at the nest for pad insertion until resumption of incubation; n = 12, range = 1.8–39.6 min). If the outlying value is excluded (because this bird was not actually on the nest when it was approached and so was likely to have been on a natural recess from incubation), then the period of disturbance was only 7.2 ± 1.2 min (n = 11, range = 1.8–13.2 min).

Does a reduction in egg heating costs lead to an increase in attendance?

We predicted that if the cost of heating eggs significantly affects a female’s ability to allocate resources to incubation, then reducing egg heating costs would increase attendance. Indeed, attendance increased significantly while nests were experimentally heated: at lower ground temperatures, attendance was considerably greater in experimental nests (Table 1 and Figure 4). The predicted value from the model (Table 1) for an experimental bird under average conditions (mean values used for ground temperature and mass reserves for the n = 28 experimental birds) is an increase in attendance of 3.6% (from 80.8–83.7% of the day, a difference of 51.8 min), but under the conditions of lowest ground temperature recorded during a sampling period (2.0°C), and mean value for mass reserves, the predicted value from the model is an increase in attendance of 14.7% (80.5–92.3%; 211.7 min). Overall attendance increased significantly for both experimental and nonexperimental birds as body mass reserves increased (Table 1 and Figure 4); for every extra 1 g of body mass, proportion of attendance increased by 0.5% daily.

When attendance (controlling for mass reserves and day of incubation) at an experimental nest was compared to a randomly chosen control nest over the same approximate 24-h sampling period on a matched-pairs basis, attendance was higher in experimentally heated nests (5.3 ± 2.2% higher; n = 7; 76.3 min): this difference was just at the level of significance despite the small sample sizes (matched-pairs t test, t_{6} = 2.4, p = .050).

Table 1
The effect of the experimental treatment of nest heating, mass reserves of the female, and ground temperature on the proportion of time spent in nest attendance

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<th>F</th>
<th>Sig.</th>
<th>β</th>
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<td>2.7</td>
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<td>Intercept</td>
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<td>5.5</td>
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<tr>
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<td>20.09</td>
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<tr>
<td>Mean ground temperature</td>
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<td>1</td>
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<td>0.044</td>
<td>0.002</td>
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<tr>
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<td>5.0</td>
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<td>Corrected total</td>
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<td>27</td>
<td></td>
<td></td>
<td>.19</td>
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</table>

The proportions of attendance were normally distributed, and so, the variable was untransformed for simplicity according to the recommendations in Sokal and Rohlf (1981). Variables removed from the model were day of incubation (F_{1,23} = 0.3, p = .53). The effect of experimental treatment (p = .010) and the interaction of treatment × ground temperature (p = .014) remained significant in this full model.

*a β values for experimental effects are given.

Figure 4
The effect of experimental heating on nest attendance showing the interaction, or not, with ground temperature (A) and mass reserves (B), plotted as mass standardized for size, i.e. {([mass/(wing length3)] × mean wing length of sample population3) g}. Residuals for the proportion of attendance were plotted after controlling for the effects of size in A and for the effects of ground temperature in B (see Table 1). Experimental females are shown as filled squares; control females, as open squares.
Table 2
The effect of nest attendance on rate of mass change (g h⁻¹)

<table>
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<tbody>
<tr>
<td>Corrected model</td>
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<td>1</td>
<td>17</td>
<td>&lt;0.001</td>
<td></td>
</tr>
<tr>
<td>Intercept</td>
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<tr>
<td>Proportion of attendance</td>
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<td>1</td>
<td>17</td>
<td>&lt;0.001</td>
<td>-1.2</td>
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<tr>
<td>Error</td>
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<td></td>
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<td>Corrected total</td>
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<td>23</td>
<td></td>
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</table>

Variables removed from the model were experimental treatment EXP (F₁₆ = 2.8, p = .11), day of incubation (F₁₆ = 0.03, p = .86), mass reserves (F₁₆ = 1.0, p = .34), ground temperature (F₁₆ = 0.2, p = .67), and EXP × ground temperature (F₁₆ = 3.8, p = .07). Proportion of attendance was significant in this full model (p = .001).

Does reducing egg heating costs reduce mass loss?

If the cost of heating eggs significantly affects a female’s energy requirements, then reducing egg heating costs may decrease rate of mass loss during attendance at the nest. The rate of mass change over the experimental period, however, was independent of experimental heating when controlling for attendance but was significantly negatively correlated with attendance (Table 2). Therefore, as birds spent more time off the nest (presumably feeding) so more mass was gained (Figure 5): for every 10% increase in daily proportion of attendance, 0.12 extra grams of body mass were lost per hour.

Does reducing egg heating costs reduce DEE?

If egg heating costs are a major component of a female pectoral sandpiper’s daily energy budget, then a reduction in the cost of heating eggs may lead to a reduction in DEE. DEE was, however, independent of experimental heating when controlling for attendance (Table 3). DEE increased significantly with mass change: as more mass was gained, DEE increased so that for every 0.1 g of mass gained per hour, DEE increased by 20.5 kJ per day (Table 3 and Figure 6).

As we could not detect any difference in DEE between experimental and control females, we estimated mean DEE across both groups combined. In pectoral sandpipers, DEE averaged 361.1 ± 8.9 kJ d⁻¹ (n = 24): 361.9 ± 13.0 for control birds and 359.8 ± 10.8 for birds on heated nests. Pectoral sandpipers therefore had a mean power output of 4.1 ± 0.1 W, equivalent to 6.1 ± 0.1 BMR (6.15 BMR for control and 6.10 for experimental birds). These values did not change significantly when alternative estimates of background concentrations of H₂¹⁸O and ²H₂O were used. For example, if background values obtained from rainwater collected at the site during June and July in the 1960s were used to calculate DEE (see Methods), either uncorrected DEE was 352.7 ± 8.6 kJ d⁻¹ or 6.0 ± 0.14 BMR, or corrected according to Tatner (1990), DEE was 372.6 ± 9.1 kJ d⁻¹ or 6.3 ± 0.15 BMR. Similarly, if background values for dipper are used (Tatner, 1990), DEE was 363.3 ± 9.0 kJ d⁻¹ or 6.2 ± 0.15 BMR; values for common sandpiper (Tatner, 1990), DEE was 361.6 ± 9.0 kJ d⁻¹ or 6.1 ± 0.15 BMR.

DISCUSSION

We determined whether the energetic costs of heating eggs constrain the behavior of female pectoral sandpipers by reducing these costs and determining whether there was any effect on (1) a female’s nest attendance, (2) change in mass, and (3) DEE. Attendance may increase, mass loss decrease, and DEE decrease, if the demand of egg heating significantly constrains a female’s energy budget. Predictions concerning nest attendance are reasonably straightforward, as birds constrained energetically from dedicating more time to incubation might respond to a reduction in costs by increasing nest attendance. Mass loss, however, can be an indication of reproductive stress or of programmed reserve depletion, so is perhaps not so easily interpreted. Changes in DEE are also difficult to predict, as a number of behavioral responses are possible, and the relative costs of component activities, particularly off the nest, were not known. For example, DEE will not change at all if time spent in an energetically demanding activity off the nest is replaced by an equally demanding activity on the nest.

We found a significant effect of reducing egg heating costs only on attendance, which increased slightly, although the effect of the cost reduction was much larger when ground temperature was lower. Although change in mass over the experimental period was determined by the proportion of overall time spent in attendance, rates of mass loss by sitting birds were unaffected by a reduction in egg heating costs. DEE was itself probably primarily determined by energy expended off the nest while birds increased in mass during feeding: more energy was used as more mass was gained, independently of the reduction in egg heating costs.

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Attendance

The result that nest attendance increased as egg heating costs decreased is consistent with the idea that an incubating
female remains on its nest until its body reserves fall to a set minimum level for that bout of incubation (see Chaurand and Weimerskirch, 1994; Cresswell et al., 2003; Weimerskirch, 1995). Then they must leave the nest to feed to replenish, or at least partly replenish, their body reserves to another set level. However, our study gives no information on the proximate cues that may initiate trips. Pectoral sandpipers pattern of incubation scheduling varies through the day, possibly in response to feeding efficiency (Norton, 1972); although the level of body reserves affects average attendance, pectoral sandpipers probably do not simply initiate trips when their body reserves drop to a critical level. If the rate of reserve utilization by a sitting bird depends at least partly on egg heating costs, then a decrease in these energetic costs will lead to a slower use of reserves and a longer period before the threshold for departure is reached. In the present study, body reserves were probably depleted at a slower rate because of the experimental nest heating, so birds on heated nests could sit for longer on average over the experimental period. This result is consistent with those of recent studies (Bryan and Bryant, 1999; Reid et al., 1999). Our result showing that birds with higher mass reserves attended more is also consistent with several other studies in which attentiveness has also been shown to depend on body condition in several species (see Aldrich and Raveling, 1983; Hegyi and Sasvári, 1998; Norton, 1973). Similarly, attendance was also probably higher when it was warmer: other studies have also shown that energetic costs during incubation depend on ambient temperature when reserves will be depleted more slowly (see Mallory and Weatherhead, 1993; Moreno, 1989a).

The interaction between ground temperature and nest heating can probably be broadly explained in terms of reserve depletion, in which particular weather conditions increased the biological significance of a reduction in egg heating costs. The effect of experimental heating was more pronounced when ground temperatures were lower when pectoral sandpipers with heated nests attended more. At average ground temperatures, attendance in experimental nests was only slightly higher than that in control nests. This suggests that attendance at low ground temperatures is more constrained: more energy will be required for heating eggs, such that reserve depletion rates will be higher resulting in shorter attendance bouts.

Overall attendance at the nest was about 82%, which is similar to other uniparental species with precocial young (Deeming, 2002). There was no effect of incubation stage on attendance (Table 1), a pattern in many species (Skutch, 1962). Initial starting mass did not affect how the experimental treatment affected attendance (mass × experimental treatment term added to the model in Table 1, \( F_{1,22} = 0.3, p = .86 \)). This is an important consideration because the costs of incubation owing to energetic limitations may only be manifest in individuals in poor condition (Heaney and Monaghan, 1996; Minguez, 1998; Moreno and Sanz, 1994).

Change in mass

Birds gained more mass if they spent longer off the nest feeding and lost more mass if they attended for longer: this result is a clear demonstration of the trade-off between incubation and foraging on body reserves that has been demonstrated in several species (see Drent et al., 1985; Tveraa et al., 1997). Mass loss during incubation bouts and over the incubation period has been shown in many species (McCluskie and Sedinger, 2000; Peters, 1983) and is inevitable for any nonforaging animal: mass loss during incubation may also be adaptive (Moreno, 1989b). There was, however, no effect of experimental heating on rate of mass change during the experimental period after controlling for attendance, even though birds with reduced costs of egg heating should have lost mass more slowly. This also, as argued above, is consistent with the reserve depletion hypothesis of attendance. If this hypothesis is correct, then both experimental and control birds will lose and regain the same amount of reserves each cycle of incubation bout and subsequent trip, although the total cycle length will be longer for experimental birds. As several cycles (on average, 22) occur in a 24-h period, then on average there will be no change in reserves recorded for experimental or control birds. There should however, have been fewer cycles for experimental birds and so less DEE.

Daily Energy Expenditure

If pectoral sandpipers with experimentally heated nests have fewer cycles of incubation and feeding (reserve loss and gain) per day, then they should use less energy per day. DEE, however, varied primarily with mass change controlling for attendance, not experimental heating. The result may suggest that variations in strategies of reserve depletion exist. The
result possibly also suggests that it is intake rate (see also Weathers and Sullivan, 1989), not the time available for feeding that limits energy intake and that there was variation in the acquisition of additional reserves independent of the experiment. Some individuals may have had higher intake rates, used more energy and gained more mass than others, independently of whether their egg heating costs were reduced. This may arise because feeding conditions may have been unpredictable, such that pectoral sandpipers fed much more intensively whenever they could, and/or because some females may have had access to better feeding areas. Experimental treatments were likely to have been random in time and space relative to local favorable feeding conditions, so any effect of reduction in energy use because of the experiment may have been swamped by the energy used when feeding. The results thus suggest that energy spent while attending the nest on at least a daily basis is less compared with those of feeding or other activities off the nest, despite the majority of time being spent on the nest. Little is known about how incubating birds and particularly uniparental incubators achieve high intake rates to compensate for their reduced foraging time, but this compensation is probably common (Tinbergen and Williams, 2002).

What then are the significant energetic costs during the incubation period?

Breeding in the arctic is clearly demanding in overall energetic terms: incubating pectoral sandpipers have a mean power output of 4.2 W, equivalent to 6.1 BMR (Kersten and Piersma, 1986), one of the highest power outputs of any bird species yet recorded, and much higher than the 2.7 BMR for incubating starlings (Ricklefs and Williams, 1984), typical of many bird species (Tinbergen and Williams, 2002). Our values are much more similar to those from other arctic breeding shorebirds. For example, two similar species (congeneric, arctic breeding, and at least occasionally uniparental)—the little stint, Calidris minuta, and red-necked stint, C. fuscirostris—have a mean daily power output while incubating equivalent to 5.6 and 4.6 BMR, respectively (Piersma et al., 2003). Biparental arctic breeding turnstouns, Arenaria interpres, have a mean daily power output of about 4 BMR when incubating (Piersma and Morrison 1994). In general, incubating arctic breeding shorebirds have an average mean daily power output equivalent to 4.2 BMR (Tinbergen and Williams, 2002), but this average based on nine species is derived mostly from biparental species. Our results, based on a relatively large sample size (see Tinbergen and Williams, 2002), clearly indicate that the DEE of some arctic incubators may be higher than previously appreciated. Such high DEEs may be possible in the arctic because food is relatively more available (either in abundance or because there is constant daylight). Nevertheless, high DEEs may have concomitant fitness costs (Daan et al., 1996).

To conclude, this experiment reveals that the costs of heating eggs can influence the length of time pectoral sandpipers can incubate before they run down their reserves, particularly when the ground is cold. However, the costs of heating the eggs may be relatively small proportion of total DEE, as costs off the nest must far exceed those incurred while sitting, even though there is relatively little time spent off the nest. Heat loss away from the favorable nest microclimate, travel to and from feeding area, and feeding costs are all probably large, swamp egg heating costs. The present study was not designed to determine whether the costs of heating eggs have fitness consequences (see Reid et al., 2000), but the results imply that pectoral sandpipers are using large amounts of energy daily during the incubation period and so may face life-history tradeoffs as a consequence. The present study is one of the few studies ever that have measured DEE with respect to an experimental manipulation and with a relatively large sample size (Williams and Vézina, 2001). Our results are therefore likely to be robust and suggest strongly that future studies of constraints on incubation behavior should concentrate on activities off the nest.

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