Foot-Mediated Incubation: Nazca Booby (*Sula granti*) Feet as Surrogate Brood Patches

Stephanie M. Morgan
Miriam A. Ashley-Ross*
David J. Anderson
Department of Biology, Wake Forest University, Winston-Salem, North Carolina 27109

Accepted 1/23/03

ABSTRACT

Incubation in most avian species involves transferring heat from parent to egg through a highly vascularized brood patch. Some birds, however, do not develop a brood patch. Unusual among birds, these species hold their eggs under the webs of their feet, but the role of the feet in heat transfer is uncertain. Often the webs are positioned between the feathered abdomen and the egg during incubation, suggesting that either the abdomen, the feet, or both could transfer heat to the egg. We studied heat transfer from foot webs to eggs during incubation in Nazca boobies by spatially separating the feet from the abdomen using an oversized egg. We found that feet transfer heat to eggs independently of any heat that may be transferred from the abdomen. In addition, we found that incubating boobies had significantly greater vascularization in their foot webs, measured as a percentage of web area covered by vessels, than nonincubating boobies. We also found that males, whether incubating or nonincubating, had significantly less web vascularization than females. We concluded that vascularized Nazca booby feet function in the same way during incubation that vascularized brood patches do, acting as surrogate brood patches.

Introduction

Incubation is loosely defined as the parental act of regulating the factors that influence embryonic development (Drent 1975; Bergstrom 1989). During incubation, one of the most important factors controlled by the parent is temperature of the developing embryo (Baldwin and Kendeigh 1932; Wilson 1991; Decuypere and Michels 1992). Extensive studies of temperature and development have shown that abnormalities and even death occur when temperature deviates from a specific range (reviewed by Romanoff 1960; Lundy 1969; White and Kinney 1974; Drent 1975; Deeming and Ferguson 1991). Most birds control changes in egg temperature using a ventrally located brood patch, typically an area of bare vascularized skin on the abdomen that is slightly thicker than surrounding skin and swollen with edema (Lange 1928; Bailey 1952). This increased vascularization seems to facilitate heat exchange between the parent and the eggs (Peterson 1955; Drent 1973; Carey 1980; Brummermann and Reinertsen 1991, 1992).

Some birds do not develop brood patches for incubation. Megapodes use mounds of decaying material to incubate their eggs (Frith 1956). Birds in the traditional order Pelecaniformes (including pelicans, boobies, gannets, anhingas, tropic birds, and cormorants) have totipalmate feet with webbing between all four toes in contrast to the palmate foot with webs between only three toes, which is typical of most other aquatic birds (Drent 1975; Hedges and Sibley 1994). Most pelecaniform birds, including boobies and gannets (Nelson 1978), do not develop a brood patch. However, they do wrap their feet around the top and sides of their eggs and crouch over them. Some have suggested that these feet transfer body heat to the egg (Howell and Bartholomew 1962; Bartholomew 1966; Drent 1975; Nelson 1978).

Heat could be transferred to and from the egg at points of contact with the abdomen, feet, air, and substrate (Fig. 1). Air and substrate temperatures in most thermal environments are lower and more variable than normal incubation temperatures, hence the need for parental heat input. This heat could come either from the abdomen and then passed through the feet to the egg by conduction, or it could be transferred directly from the feet by conduction.

To determine whether feet added heat to eggs independently of heat from the abdomen, Howell and Bartholomew (1962) measured foot temperatures in incubating red-footed boobies (*Sula sula*) by folding the webs of their totipalmate feet around a thermistor until a temperature reading could be obtained. Boobies incubate with the two medial webs of each foot wrapped around the egg. Howell and Bartholomew (1962) found no difference between foot and egg surface temperatures and concluded that the egg's heat originated at the abdominal surface, passing through the feet where the feet were sandwiched between the abdomen and egg. In their view, the feet did not constitute an independent source of heat flow into the egg; instead, the feet conducted heat from the abdominal sur-

---

* Corresponding author; e-mail: rossma@wfu.edu.

Physiological and Biochemical Zoology 76(3):360–366. 2003. 1522-2152/2003/7603-2122$15.00

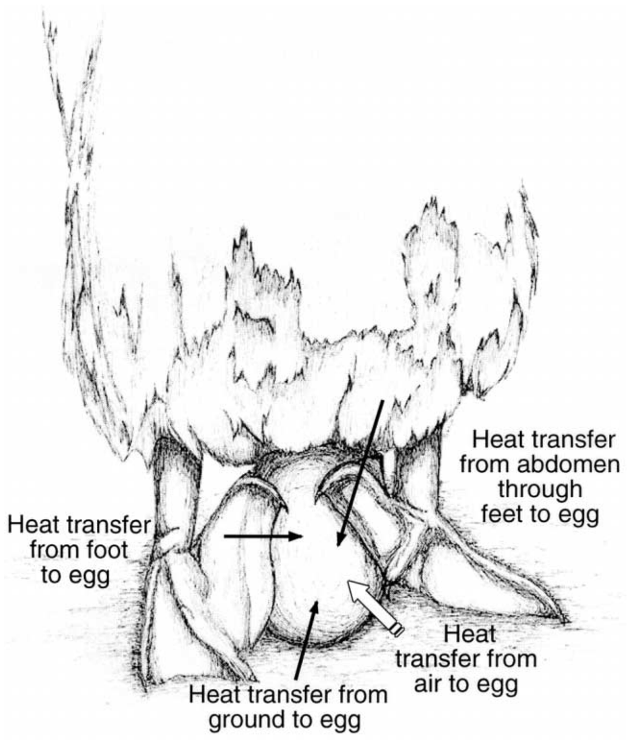

Figure 1. Possible avenues for heat transfer to the egg in foot-mediated incubation. Arrows represent heat flow, as indicated.

face to the egg or conducted heat from the egg to the environment (Fig. 1).

Nelson (1978), reviewing similar work by Howell (unpublished data cited in Nelson 1978) on blue-footed boobies (*Sula nebouxii*), rejected this conclusion, suggesting that the result was an artifact of the manner in which the data were taken. He pointed out that holding the thermistor within the fold of the foot web through the skin in the foot could have caused vasoconstriction and lowered foot temperatures and that the time elapsed between capturing birds and measuring foot temperatures could have allowed the feet to cool significantly. Others have left the debate as unresolved (Drent 1975; Evans 1995; Whittow 2002), saying that heat could pass from the abdomen through the feet as Howell and Bartholomew (1962) claimed.

We studied the role of the feet during incubation in Nazca boobies (*Sula granti*; Sulidae), known until recently as a subspecies of masked boobies (*Sula dactylatra*; American Ornithologists' Union 2000). We tested the hypothesis that the feet transfer heat from the parent to the eggs independently of any heat transfer that may occur from the abdominal surface to the eggs. We recorded temperatures of the feet and egg when the feet were spatially separated from the abdomen during incubation. We also examined the likely mechanism of heat transfer to eggs by comparing the degree of vascularization in foot webs of incubating and nonincubating boobies.

Methods

We studied Nazca boobies nesting on the island of Española, Galápagos, Ecuador (1°20′S, 89°40′W; see Anderson and Ricklefs 1987) during the 1999–2000 (vascularization measurements) and 2000–2001 (heat exchange measurements) breeding seasons. Nazca boobies lay one or two eggs in a scrape on the ground (Anderson 1993) and readily tolerate approach and handling while incubating (Nelson 1978).

Foot Heat Exchange

When incubating, boobies normally wrap both feet around the sides and top of their eggs and crouch over their eggs, positioning some of the webbing between the abdomen and the eggs (Nelson 1978; Anderson 1993; Fig. 2*A*). We separated the feet from the abdomen by substituting an abandoned waved albatross (*Phoebastria irrorata*) egg for the natural eggs in four two-egg clutches. The albatross eggs are approximately twice as long and twice as wide as natural Nazca booby eggs (Fig. 2*B*). When an adult booby incubated an albatross egg, its webbed feet were displaced to the sides of the albatross egg and were not in contact with the abdomen touching the upper surface or with each other (Fig. 2*C*).

We used intact waved albatross eggs, abandoned during the previous season in their nearby nesting colony (Harris 1973). We drilled small holes in the ends of the albatross eggs, blew out the contents, and filled them with water. We then sealed the holes and attached three thermistor probes to the egg with duct tape (Fig. 2*B*, 2*C*). Two thermistors were positioned, one on either side of the egg, to assure full contact with each foot. The third thermistor on the top rear of the egg touched the abdomen, but not the feet, during incubation. The probes led from the back of the egg to a data logger (HOBO 4-channel external temperature data logger, Onset Computer Corporation, Bourne, Mass.) outside the nest scrape. A small arrow on the end of the egg indicated the position of the abdomen probe during incubation. In all four trials, the arrow remained vertical, showing that the egg was positioned as we intended.

Incubating adults (two male, two female) were chosen at random in the mornings between 0730 hours and 0930 hours when the ground and air temperatures were uniform and low. The adult was removed briefly from the nest, and the temperatures of the upper surface of its own egg and of the albatross egg were taken using an infrared sensor (Raynger ST20 Pro standard, Raytek Corporation, Santa Cruz, Calif.). We replaced the original booby eggs with the modified albatross egg, and the adult returned to the nest and incubated it. Original booby eggs were maintained at incubation temperature during the trial, and an observer watched the incubating adult from 4–8 m away. In all cases, the adult accepted the albatross egg and incubated it with its feet in the intended lateral positions. Because the albatross egg temperature equaled the low ambient

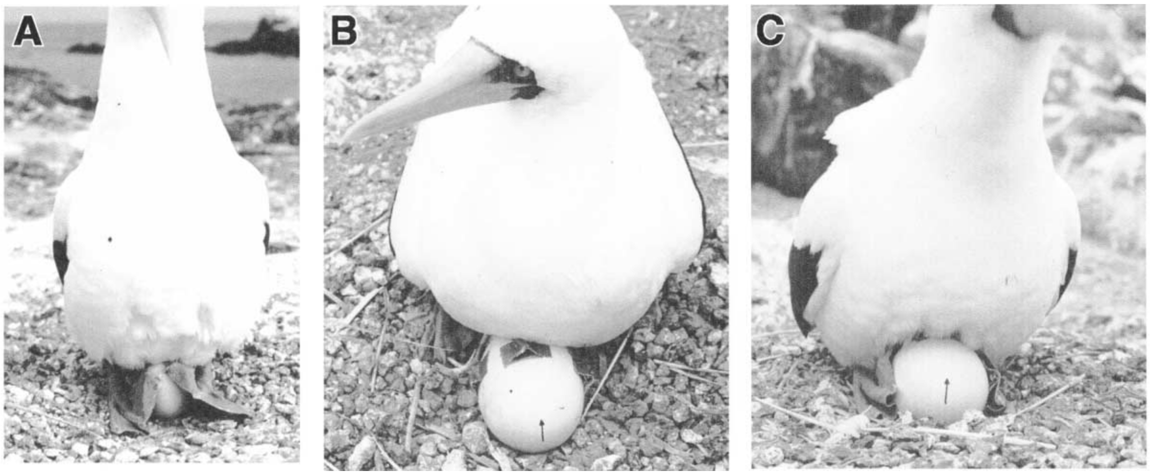

Figure 2. Incubation posture of Nazca booby during incubation of its own egg and the modified albatross egg. *A*, Nazca booby incubating its own egg. The bird is slightly raised, exposing the position of the feet. *B*, Nazca booby preparing to incubate a modified albatross egg; the bird is slightly raised from normal. Gray duct tape covers two temperature sensors on either side of the egg that correspond to the position of the feet during incubation; the third sensor at the top corresponds to the position of the abdomen during incubation. *C*, Nazca booby incubating a modified albatross egg. The feet can be seen covering their respective temperature probes while feathers obscure the abdomen probe. The arrow at the end of the egg allows the observer to confirm that the abdomen is in the correct position. Note that the abdomen does not cover the feet as in *A*, due to the size of the egg relative to the bird.

temperature (24°–26°C) at the start of each trial, the experiment challenged adults to warm a large, cool egg. Air temperatures from a shaded area were monitored throughout each trial with a temperature data logger (StowAway TidbiT, Onset Computer Corporation, Bourne, Mass.), and ground temperatures under the eggs were measured with the infrared sensor at the end of each trial.

Trials lasted 19 to 110 min, until the adult attempted to turn the albatross egg. At that point, the bird was removed, and the temperature of the warmed albatross egg was taken again using the infrared sensor before it was replaced with the parent's original egg. This procedure was repeated on four mornings at four different nests.

Foot Vascularization

Sunlight passing through Nazca booby foot webs illuminated their vascularization. During February and March 2000, 17 incubating male and 18 incubating female boobies were removed from their nests between 0730 hours and 0930 hours, and their feet were videotaped with the sunlight behind them. The left and right foot of each bird were videotaped separately. This procedure was repeated with 18 nonincubating males and 19 nonincubating females during the same time period. Nonincubating birds were defined as adult birds standing at or near nests in the colony but with no eggs or chicks at the moment. The birds may have had a nest at some point earlier in the season; thus, they could have been incubating eggs at some point in the weeks before the study.

Still images of each individual's left and right foot were captured from the video using Image Pro Plus (Media Cybernetics, Silver Spring, Md.). We examined the degree of vascularization in the middle web of each foot because it is one of the webs spread around the eggs during incubation (Bartholomew 1966) and because it was the web most consistently held perpendicular to the focal axis of the camera by the birds. To more accurately characterize the web's vascularization, images were manipulated to increase brightness and contrast and then sharpened. In order to avoid biasing results, one image of one foot (either right or left) was chosen for the analysis by an outside volunteer and assigned a random number.

Still images named as random numbers were opened in Adobe Photoshop (Adobe Systems, San Jose, Calif.) and manipulated to enhance the contrast between the vascularization and webbing. We used Image Pro Plus to select the webbing and then used the histogram function in Photoshop to determine the proportion of pixels showing vascularization. We transformed the proportions using an arcsine transformation described by Zar (1984) and then used the transformed numbers in a two-way ANOVA in StatView (SAS Institute, Cary, N.C.).

Results

Foot Heat Production

The temperature of the albatross egg increased throughout all four trials (Fig. 3): three of four incubating adults shivered

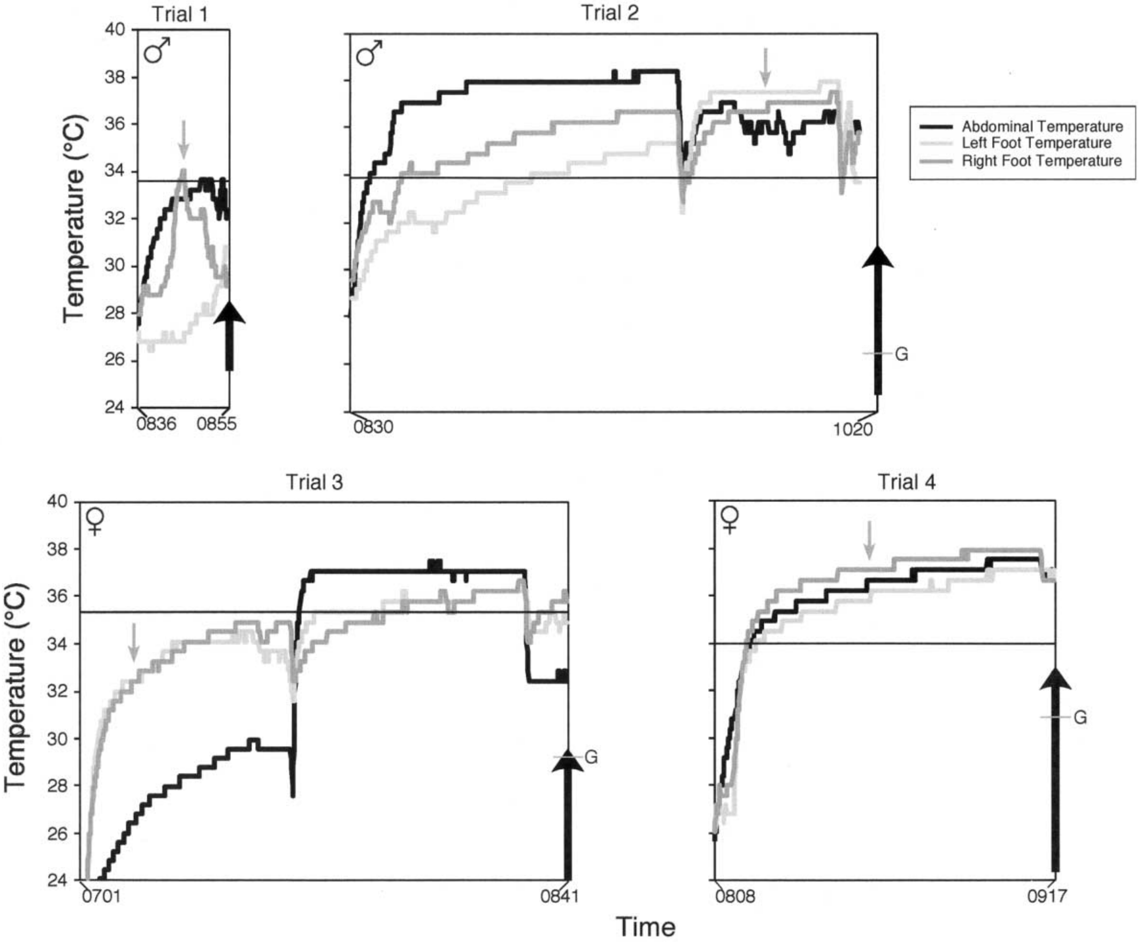

Figure 3. Plots of the temperatures of Nazca booby feet and abdomen temperatures during incubation of a modified albatross egg. Each panel represents a different trial and different individual bird (male or female, marked by symbols within each panel). The width of the *X*-axis varies with the length of the trial. The *Y*-axis is the same temperature scale for all panels. The horizontal line in each panel is the temperature of the original booby egg taken with an infrared sensor before being replaced with the modified albatross egg. The large arrow on the right of each panel represents the change in temperature of the albatross egg over the length of the trial: the bottom of the arrow corresponds to the temperature of the egg taken with the sensor before the bird was allowed to incubate, the top of the arrow corresponds to the temperature of the egg after the bird was removed but before its own egg was replaced in the nest, and the gray line (*G*) crossing the arrow indicates the ground temperature at the end of the trial. Dark gray lines represent the right foot temperature, light gray lines the left foot temperature, and thick black lines the abdomen. Small gray arrows in the panels indicate periods during which one or both feet were warmer than the abdomen. The final albatross egg temperature in all trials exceeded the maximum ambient temperature (26.6°C). Ground temperature was recorded under the bird at the end of three of the four trials (trial 2, 26.3°C; trial 3, 29.2°C; trial 4, 30.9°C).

during this process. Albatross egg temperatures at the end of all trials exceeded 28.0°C, while ambient temperatures never exceeded 26.6°C. Final egg temperature likewise always exceeded final ground temperature under the egg (Fig. 3; ground temperature was not measured in trial 1). Thus, heat flow throughout the trials was apparently from neither the air nor the ground into the eggs. Temperatures recorded from one or both of the feet exceeded the temperature of the abdomen at some point during each trial (Fig. 3). Temperatures of the feet exceeded the final temperature of the albatross egg throughout most of each trial and, in fact, often exceeded the original temperature of the booby egg. Separate experiments where the feet were painted with temperature-sensitive paint showed that the feet of incubating Nazca boobies reached temperatures above 40°C (S. M. Morgan, unpublished data).

The duration of each trial (Fig. 3) varied since individuals differed in the length of time they incubated before attempting to turn the albatross egg. Spikes or sudden drops in temperature corresponded to our observations of the bird repositioning the egg (especially in the abdomen temperature; Fig. 3), indicating that at some points the bird was not always sitting tightly on all three probes.

Foot Vascularization

The relative area of blood vessels (Fig. 4) in incubating birds' feet exceeded that of nonincubating birds in both males and females (Fig. 5; ANOVA incubator-nonincubator effect, $F_{1,70} = 8.407$, $P = 0.005$). The relative area of vessels in female birds' feet exceeded that of males (Fig. 5; ANOVA sex effect, $F_{1,70} = 6.449$, $P = 0.01$). The interaction between sex and incubation status was not significant ($F_{1,70} = 1.072$, $P = 0.30$).

Discussion

During incubation when birds cover their eggs with their feet, several potential routes of heat flow into the egg exist (Fig. 1). As discussed earlier, heat could flow from the ground or the air into the egg, given the appropriate thermal gradient. Heat could also pass from the adult's abdomen through the feet and into the egg. This route is the one advocated by Howell and Bartholomew (1962) and some other authors (Drent 1975; Evans 1995; Whittow 2002). In addition, heat could pass from the adult's foot webs to the eggs without involvement of the abdominal surface.

In the trials described here, ambient air and ground temperatures were equal to or below the final albatross egg temperatures, meaning that heat did not flow into the egg by either route (the assumption that ambient temperature warmed throughout each trial was supported by the air temperature data, so the final air and ground temperatures represent the maxima during the trials). Nonetheless, egg temperature increased during each trial, so heat from the incubating adult clearly flowed into the egg. During most of each trial, the temperature of one or both feet exceeded the final temperature of the albatross egg after warming. Assuming that the final egg temperature was its maximum temperature, heat flowed from the feet into the egg and not the reverse. Furthermore, the foot webs reached temperatures higher than the abdomen at some point in each trial (Fig. 3), indicating that at those times, at least, the temperature gradient from the foot to the egg was steeper than that from the abdomen to the egg.

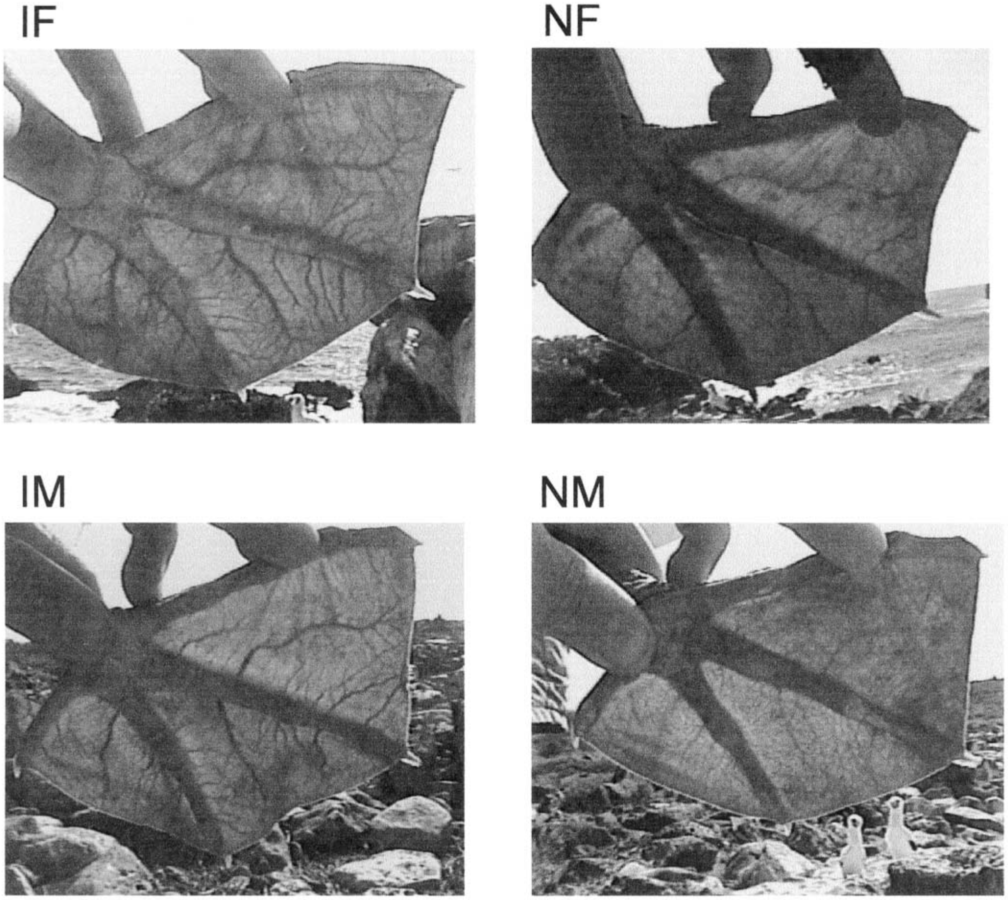

Figure 4. Vascularization in feet of four Nazca boobies. *IF* = incubating female; *NF* = nonincubating female; *IM* = incubating male; *NM* = nonincubating male. Incubating birds have more vascularization than nonincubating birds.

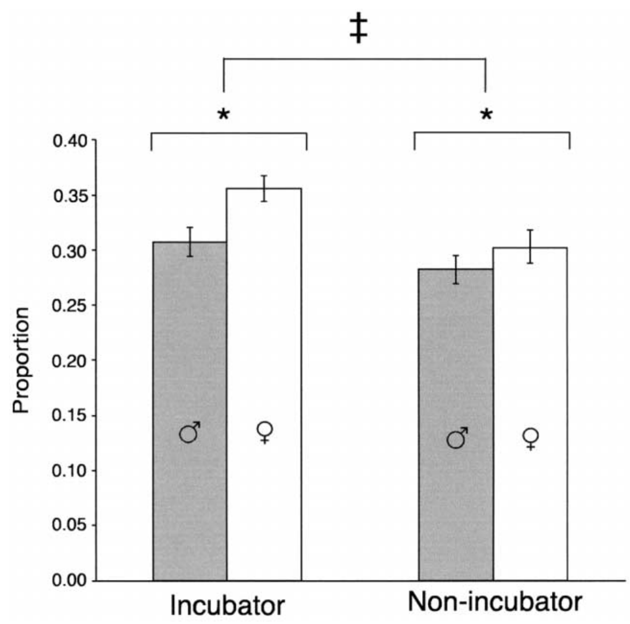

Figure 5. Comparison of the proportion of vascularization in incubating and nonincubating foot webs. Incubating Nazca boobies have significantly more vascularization in their foot webs than nonincubating Nazca boobies. Females have significantly more vascularization than males, but there is no interaction between sex and incubation status (brackets with symbols indicate statistically significant differences).

Foot temperatures exceeded albatross egg temperature during trials and also exceeded the temperature of the original booby egg in each trial (Fig. 3). These data clearly indicate that feet are capable of adding heat to eggs during incubation. Most of the adults' feet reached temperatures above the final albatross egg temperature within the first 10 min of the trial, which suggests that parents can increase heat output in reaction to cool egg temperatures. All of these results directly contradict the conclusions of Howell and Bartholomew (1962), who recorded foot temperatures at or slightly below egg incubation temperatures. However, they recorded foot temperatures from incubating adults as well as from adults brooding young, and data from brooders may have lowered mean foot temperature. Additionally, they did not record temperatures under the foot web during incubation but only from within a fold of a web, which could have led to the measurement of cooler than normal temperatures (Nelson 1978).

Our findings do not, however, completely refute Howell and Bartholomew (1962). Nazca boobies, in their natural incubation position with the abdomen resting on top of parts of the feet (Fig. 2*A*), could still passively transfer heat to their eggs from their abdomen through their feet as proposed. Passive transfer, however, would be in addition to the transfer of heat from feet directly, as we have shown is possible.

For other bird species, increased vascularization of the brood patch occurs when heat transfer is required for incubation (Drent 1973; Brummermann and Reinertsen 1991, 1992). Our observations of booby feet in the wild showed that incubators have more vascular tissue in their webs than nonincubators do and suggested that they have more blood flow through their foot webs (Fig. 4). Even though we cannot determine from our data whether the number of veins and arteries increased or whether their size increased, either would presumably facilitate heat transfer to the eggs. Both an increase in size and number of vessels is known to occur in brood patches of other birds (Lange 1928; Bailey 1952).

We also demonstrated an unexpected difference in foot web vascularization between males and females (Figs. 4, 5) that was consistent between incubators and nonincubators. In general, brood patch development is believed to be associated directly with the role the adult plays in incubation; therefore, the sex that incubates less should have a less developed brood patch (Bailey 1952; Jones 1971; Skutch 1976). Indeed, Wiebe and Bortolotti (1993) found that male American kestrels (*Falco sparverius*) had smaller brood patches than females did and incubated less than females did. Male Nazca boobies have been presumed to participate equally in incubation (Nelson 1978) and would be expected to have brood patches (foot webs) similar to females, but our data refute this expectation. Our heat exchange trials demonstrate that both males and females are capable of adding heat to the egg using their feet, suggesting that the difference is not functionally significant. Skutch (1976) claims that males that participate in incubation do not necessarily have to develop a brood patch, a conclusion which Zann and Rosetto (1990) later supported in their study of male zebra finches. Whether males often develop brood patches smaller or less vascularized than females is unknown and relatively unstudied; therefore, the fact that Nazca booby males have slightly less vascularized feet than females may not be unusual.

In this study, we found support for the hypothesis that Nazca boobies use their feet as surrogate brood patches by independently adding heat to their eggs during incubation through the webs. The feet may also merely passively direct heat from the abdomen to the eggs as well. "Active" heat transfer from the feet to the eggs is likely facilitated by increased vascularization of the foot webs.

Acknowledgments

We thank the Galápagos National Park Service for permission to work in the park and the Charles Darwin Research Station and TAME airline for logistical support. National Science Foundation grant DEB96-29539 to D.J.A. and Wake Forest University provided financial support along with the Vecellio Fund of Wake Forest University. We thank H. Townsend, P. Kappes, and

L. Clifford for assistance in the field; M. T. Morgan for artwork; and M. F. Winterrowd for editing and revisions.

Literature Cited

American Ornithologists' Union. 2000. Forty-second supplement to the AOU check-list of North American Birds. Auk 117:847–848.

Anderson D.J. 1993. Masked Booby *(Sula dactylatra)*. In A. Poole and F. Gill, eds. The Birds of North America. No. 73. American Ornithologists' Union, Philadelphia.

Anderson D.J. and R.E. Ricklefs. 1987. Radio-tracking masked and blue-footed boobies (*Sula* spp.) in the Galápagos Islands. Natl Geogr Res 3:152–163.

Bailey R.E. 1952. The incubation patch of passerine birds. Condor 54:121–136.

Baldwin S.P. and S.C. Kendeigh. 1932. Physiology of the temperature of birds. Sci Publ Cleveland Mus Nat Hist 3:1–196.

Bartholomew G.A. 1966. The role of behavior in the temperature regulation of the masked booby. Condor 68:523–535.

Bergstrom P.W. 1989. Incubation temperatures of Wilson's plovers and killdeers. Condor 91:634–641.

Brummermann M. and R.E. Reinertsen. 1991. Adaptation of homeostatic thermoregulation: comparison of incubating and non-incubating Bantam hens. J Comp Physiol B 161: 133–140.

———. 1992. Cardiovascular responses to thoracic skin cooling: comparison of incubating and non-incubating Bantam hens. J Comp Physiol B 162:16–22.

Carey C. 1980. The ecology of avian incubation. BioScience 30: 819–824.

Decuypere E. and H. Michels. 1992. Incubation temperature as a management tool: a review. World's Poult Sci J 48:28–38.

Deeming D.C. and M.W.J. Ferguson. 1991. Physiological effects of incubation temperature on embryonic development in reptiles and birds. Pp. 147–171 in D.C. Deeming and M.W.J. Ferguson, eds. Egg Incubation: Its Effects on Embryonic Development in Birds and Reptiles. Cambridge University Press, Cambridge.

Drent R.H. 1973. The natural history of incubation. Pp. 262–322 in D.S. Farner, ed. Breeding Biology of Birds. National Academy of Sciences, Washington, D.C.

———. 1975. Incubation. Pp. 331–420 in D.S. Farner, J.R. King, and K.C. Parkes, eds. Avian Biology. Vol. 5. Academic Press, New York.

Evans R.M. 1995. Incubation temperature in the Australasian gannet *Morus serrator*. Ibis 137:340–344.

Frith H.J. 1956. Breeding habits in the family Megapodiidae. Ibis 98:620–640.

Harris M.P. 1973. The biology of the waved albatross *Diomedea irrorata* of Hood Island, Galápagos. Ibis 115:483–510.

Hedges S.B. and C.G. Sibley. 1994. Molecules vs. morphology in avian evolution: the case of the "pelecaniform" birds. Proc Natl Acad Sci USA 91:9861–9865.

Howell T.R. and G.A. Bartholomew. 1962. Temperature regulation in the red-tailed tropicbird and the red-footed booby. Condor 64:6–18.

Jones R.E. 1971. The incubation patch of birds. Biol Rev 46: 315–339.

Lange B. 1928. Die Brutflecke der Vögel und die für sie wichtigen Hauteigentumlichkeiten. Gegenbaurs Morphol Jahrb 59:601–712.

Lundy H. 1969. A review of the effects of temperature, humidity, turning and gaseous environment in the incubator on the hatchability of the hen's egg. Pp. 143–176 in T.C. Carter and B.M. Freeman, eds. The Fertility and Hatchability of the Hen's Egg. Oliver & Boyd, Edinburgh.

Nelson J.B. 1978. The Sulidae: Gannets and Boobies. Oxford University Press, Oxford.

Peterson A.J. 1955. The breeding cycle in the bank swallow. Wilson Bull 67:235–286.

Romanoff A.L. 1960. The Avian Embryo. Macmillan, New York.

Skutch A.F. 1976. Parent birds and their young. University of Texas Press, Austin.

White F.N. and J.L. Kinney. 1974. Avian incubation. Science 186:107–115.

Whittow G.C. 2002. Seabird reproductive physiology and energetics. Pp. 409–437 in E.A. Schreiber and J. Burger, eds. Biology of Marine Birds. CRC, New York.

Wiebe K.L. and G.R. Bortolotti. 1993. Brood patches of American kestrels: an ecological and evolutionary perspective. Ornis Scand 24:197–204.

Wilson H.R. 1991. Physiological requirements of the developing embryo: temperature and turning. Pp. 145–156 in S.G. Tullet, ed. Avian Incubation. London Poultry Science Symposium 22. Butterworth-Heinemann, London.

Zann R. and M. Rossetto. 1990. Zebra finch incubation: brood patch, egg temperature and thermal properties of the nest. Emu 91:107–120.

Zar J.H. 1984. Biostatistical Analysis. 2d ed. Prentice Hall, Englewood Cliffs, N.J.