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MEASURING RESTING HEART RATES IN PENGUINS USING AN ARTIFICIAL EGG

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Abstract.—We report on the use of artificial eggs, fitted with highly sensitive microphones to record the resting heart rates of Adélie Penguins (*Pygoscelis adeliae*). Artificial eggs were developed to quantify the effects of recording heart rate in penguins using electrocardiogram (ECG) units attached externally to the penguins. We found the resting heart rates of penguins recorded using artificial eggs were indistinguishable from those recorded using ECG units. Artificial eggs are a useful tool for quantifying the effects of other, more invasive techniques such as attaching ECG units. Artificial eggs are also potentially useful in physiology and energetics studies because they are simple to use, are relatively inexpensive to construct and operate, and are comparatively non-invasive to the birds. Nevertheless, some limitations of the operation and application of artificial eggs are discussed.

MIDIENDO RITMOS CARDIACOS DE PINGÜINOS (SPHENISCIFORMES) EN REPOSO UTILIZANDO UN HUEVO ARTIFICIAL

Sinopsis.—Reportamos et uso de huevos artificiales, insertados con micrófonos altamente sensitivos para registrar los ritmos cardiacos en reposo de *Pygoscelis adeliae*. Los huevos artificiales se desartollaron para cuantincar los erectos de medir el pulso cardiaco en estas aves utilizando unidades de electrocardiograma (EKG) adheridas externarnente a las aves. Hallamos que los ritmos cardiacos de la especie en reposo registrados utilizando los huevos artificiales no se distinguieron de los registrados utilizando las unidades EKG. Los huevos artificiales también son potencialmente átiles en estudios básicos de fisiologia y de energética, ya que son fáciles de usar, relativamente baratos de hacer y de operar, y son no son en lo mínímo invasivos a las aves. De todas formas, se discuten algunas limitaciones a la operación y uso de los huevos artificiales.

Concern over the potential for scientific techniques to harm animals and bias the results of research has lead to some novel developments in the collection of biological information from free-living animals (e.g., Howey et al. 1984, Ellis et al. 1991, Nimon et al. 1996). In our research on the effects of human activity on penguins (Giese 1996, 1998), techniques that reduce any confounding effects of investigator disturbance are particularly valuable, because it is vital that the effects of monitoring procedures do not mask any effects of the human activity of interest.

During a study of the effects of controlled pedestrian approach on Adélie Penguins (*Pygoscelis adeliae*), we employed external electrocardiogram (ECG) units to record penguin heart rate as an index of disturbance (M. Giese, unpubl. data). External ECG's have proven to be a reliable and versatile means of recording heart rate in Adélie Penguins (Culik et al. 1989) and require considerably less manipulation of the birds than implanting internal units (Culik 1992). Even so, to fit penguins safely with external ECG's, birds must be removed from their nests and restrained for up to 5 min while units are glued to the penguins' backs and electrode safety pins inserted subcutaneously into the penguins' chest. The potential for this procedure to modify the birds' subsequent behavior and physiology needed to be quantified in our study because we wanted to be certain we were recording responses to controlled pedestrian approaches rather than to our monitoring procedures. Therefore we developed an artificial egg as a relatively non-invasive technique for recording resting heart rate in the penguins that would enable comparisons of heart rate between birds that had been handled and fitted with ECG units and unhandled birds that wore no external devices.

We know of only one other, similar device for measuring heart rate in nesting penguins (Nimon et al. 1996). However, unlike their unit, the artificial eggs described here allow normal egg turning behavior and do not require elaborate methods of placement, such as burial and camouflage of supporting boards and wiring. More importantly, the deployment of our artificial eggs does not result in the temporary departure of incubating birds from their nests (cf. Nimon et al. 1996).

METHODS

Field procedures.—Artificial eggs were used in penguin colonies within the Prydz Bay area, east Antarctica (68°36′S, 77°58′E) during the 1993–1994 Austral summer. The units were introduced into 10 nests of incubating penguins breeding at the periphery of larger colonies (approximately 500 pairs) by balancing the artificial egg on a spoon taped to the end of a three meter pole. A second pole was held in front of the penguin to distract the bird from pecking at the unit. Once penguins had settled on the artificial eggs and re-commenced incubation (average time of 141.3 ± 54.5 s from placement), all humans moved out of sight and the birds' heart rate and behavior were recorded onto a video from within an observation hide located 30 m from the nest.

We only used nests that contained either one or no natural eggs (Adélie Penguins normally lay two eggs, Williams 1995), which meant there was no displacement of real eggs. Placement of the artificial eggs took less than 2 min. Eggs remained in the nests for up to 2 h and were removed using the modified pole.

Construction and operation of eggs.—Egg shells were constructed of an acrylic plastic sheet, which was heated and pressed over a wooden mould of an Adélie Penguin egg. The egg was constructed in two halves, at a total cost per egg of approximately U.S. $20. Eggs were fitted inside with a commercially available FM bug plus batteries and had an external antenna (Fig. 1). The FM bug was a simple frequency modulated transmitter that operated in the commercial FM radio band (88–108 Mhz), which meant that any domestic FM receiver could pick up the signal. The bugs were purchased in kit form from an electronic hobby supplier (Talking Electronics, 35 Rosewarn Ave. Cheltenham, 3192, Victoria, Australia) and required minimal electronic construction skills to assemble. The circuit board for the FM bugs we used measured 45 mm × 12 mm and cost approximately U.S. $15.

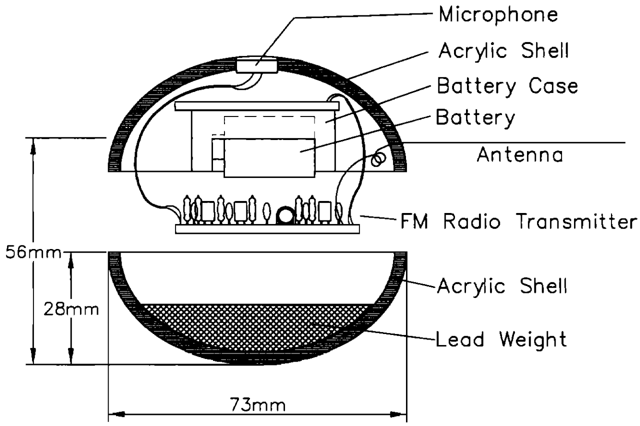

FIGURE 1. Cross section of an artificial egg with microphone to measure heart rate in Adélie Penguins.

An electec microphone was used, which was quite robust with the sensitivity being adjusted via a resistor to optimize the signal to noise ratio. The units utilized one 3.6 V 2/3 AA lithium thionyl chloride battery. The antenna was a length of wire cut to approximately a quarter wavelength of the transmitter frequency and in our eggs was 50-cm long.

The units were constructed by gluing an FM bug inside the acrylic shell. A hole was drilled into the top of the shell and the microphone flush mounted with the outside of the egg to achieve optimal acoustic coupling. The battery was tacked in place using double-sided tape to facilitate battery replacement. A reed relay switch was incorporated so that the unit could be turned off when not deployed.

The eggs had a stethoscopic effect whenever the microphone was in contact with the highly vascularized brood patch of the penguin, and this occurred during normal incubation behavior. To facilitate the microphone remaining in an up-ward facing position, but to obviate the need to secure eggs to an artificial surface within the nest (Nimon et al. 1996), our eggs were differentially weighted with lead so that if rolled by the incubating bird they would settle with the microphone facing toward the penguins' brood patch.

RESULTS AND DISCUSSION

All attempted egg placements were deemed successful, in that no eggs were rejected, no adult birds were even temporarily displaced from nests, no damage was done to real eggs, and records of resting heart rate were achieved from all trials. Penguins periodically rolled and rearranged the artificial eggs (cf. Nimon et al. 1996), and were highly protective of eggs when we went to retrieve them. These observations are indicative of the high level of acceptance of the artificial eggs by incubating penguins. In a recent study, similar, though smaller, artificial eggs were placed in Antarctic Fulmar (*Fulmarus glacialoides*) and Cape Petrel (*Daption capense*) nests with the same high degree of acceptance (M. Giese, unpubl. data). If appropriate care is taken to minimize disturbance when moving around nests and when placing eggs, other species known to display strong incubation drives and nest sight fidelity could also be expected to accept eggs unconditionally.

Penguin heart rates were able to be recorded as soon as birds settled on the eggs. Mean resting heart rates recorded from Adélie Penguins using our artificial eggs ranged from 69.5–91.7 beats/min (bpm, n = 10 penguins). These heart rates were recorded when the birds were lying prone over their eggs in a relaxed posture (on average 30.3 ± 5.8 min/2h). With behavior held constant this way, the average resting heart rates recorded from these birds (82.4 ± 8.1 bpm) were statistically indistinguishable from the resting rates of Adélies recorded using external ECG units (82.4 ± 11.7 bpm, n = 17), providing penguins were left to acclimate to wearing the ECG units for approximately 20 h (t-test = 0.03, modified df = 25, P = 0.97, statistical power = 99%). Degrees of freedom were modified in this test to account for heterogeneity of variances (Watson and McGaw 1980). The statistical power (99%) associated with this result reflects a high probability that differences in heart rate as measured by the two procedures would have been detected had they been present. The resting heart rates recorded using artificial eggs during this study were also within the range of other published accounts for Adélie Penguins recorded from external (86 ± 5 bpm, Culik et al. 1989) and internal ECG devices (73 bpm, Culik 1994).

Although artificial eggs were capable of recording the resting heart rates of Adélie Penguins during this study, their operation can be problematic. Cheaper FM bugs can drift in frequency, which can limit the number of units deployed simultaneously and the artificial eggs can be subject to local electrical interference. Moreover, because artificial eggs, as with other similar applications (e.g., Ellis et al. 1991, Nimon et al. 1996), rely on contact with the birds' brood patch to record heart rate, their application can be somewhat specific. It is impossible, for example, to record the heart rates associated with standing postures and even slight movement by the incubating adult can interfere with signal strength. Furthermore, the units are most useful during the incubation phase when adult birds are more likely to accept eggs unconditionally.

For these reasons, it has not been possible to record the heart rate responses of Adélie Penguins to human approaches using the artificial eggs. To this end, the complimentary use of ECG units has been essential in our work since they can provide a record of heart rate irrespective of the penguins' behavior or breeding phase. To illustrate, in response to a single human approaching slowly and standing 5 m from incubating Adélies, we found that 63% of the birds initially stood up (n = 30 penguins), thus exposing their eggs. These birds had not been previously handled or directly approached by humans. If artificial eggs had been used to record heart rate in these birds, the heart rate records would only be obtained from the 37% of penguins that remained lying down while approached. This would have provided an unrepresentative sub-sample of the population, or recordings would have been interrupted until birds resettled on their nests, which would give a partial, and potentially biased, sample of the bird's total behavioral response.

Nevertheless, the artificial eggs have a number of important benefits over alternatives for measuring pulse rates of free-living penguins. First, they are relatively inexpensive to construct and operate. Second, they are relatively non-invasive to the birds compared to some other techniques (e.g., Culik et al. 1989, Culik 1992) so they can provide an instant record of the heart rate response of an un-encumbered, unhandled animal. Third, the method of deployment is simple and does not displace incubating penguins from the nest nor impede the birds' normal egg-rolling behavior (e.g., Nimon et al. 1996).

To date, we have successfully trialed artificial eggs on three Antarctic seabird species; Adélie Penguins, Antarctic Fulmars, and Cape Petrels. Egg size is only limited by the sizes of FM bugs and batteries, which can be customised to suit the needs of specific applications. Deployment procedures also can be modified to suit the habitat and disposition of the species involved. For example artificial eggs were deposited into Antarctic Fulmar and Cape Petrel nests by hand.

In our work on the effects of human approach on penguins, artificial eggs were used as a complementary procedure to determine what, if any, measurable impact was associated with using ECG units. This is perhaps one of the most valuable applications of the artificial eggs, particularly given the increasing need to address issues of experimental bias arising from standard methodologies. However, artificial eggs are also appropriate for base-line physiological and energetics studies and may be particularly useful with species for which conducting more invasive procedures is restricted for biological, ethical or administrative reasons.

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