# SEXUAL DIMORPHISM IN THE PROVIDENCE PETREL Pterodroma solandri USING DNA ANALYSIS

## ADAM J. BESTER<sup>1,3</sup>, N. I. KLOMP<sup>1</sup> and D. PRIDDEL<sup>2</sup>

<sup>1</sup>Charles Sturt University, PO Box 789, Albury, NSW 2640, Australia. <sup>2</sup>Department of Environment and Conservation (NSW), PO Box 1967, Hurstville, NSW 2220, Australia. <sup>3</sup>Current address: Arid Recovery, PO Box 150 Roxby Downs, SA, 5725, Australia.

#### Received: 25 October 2005

The Providence *Petrel Pterodroma* solandri was studied on Lord Howe Island during the 2000 breeding season. Molecular sexing was conducted on feather samples taken from 18 pairs of breeding birds. Morphological measurements taken from these sexed birds indicate that males were significantly heavier than females (mean mass of males  $507.2 \pm 41.5$  g; females  $476.6 \pm 44.7$  g) and had a significantly longer culmen (males  $36.5 \pm 1.1$  mm; females  $35.5 \pm 1.3$  mm). This study confirms earlier evidence, based on small sample sizes, that Providence Petrels are indeed sexually dimorphic. Sexual dimorphism has only been positively identified in two other *Pterodroma*. The ability to sex individual Providence Petrels using this DNA analysis provides a means to investigate sex-specific differences in foraging and breeding behaviour.

### **INTRODUCTION**

Lord Howe Island and a small island off Norfolk Island (Philip Island) support the only known breeding populations of the Providence Petrel *Pterodroma solandri* in the world (Fullagar *et al.* 1974); consequently the species is listed as vulnerable under both the 2000 IUCN Red List of Threatened Animals (Criteria D2) and the New South Wales Threatened Species Conservation Act 1995, although it is not listed under commonwealth legislation (Environment Protection and Biodiversity Conservation Act 1999). On Lord Howe Island, Providence Petrels breed predominantly on Mt Lidgbird and Mt Gower, with smaller populations occurring at lower elevations on the southern section of the island (Fig. 1).

Adult body mass among the *Pterodroma* is highly variable, ranging from 163 grams for Pycroft's Petrel *P. pycrofti* to 525 grams for the Great-winged Petrel *P. macroptera macroptera* (Warham 1990). The Providence Petrel is at the heavier end of the scale, although prior to this study, morphometric data were scant and based on small sample sizes. Data provided in Marchant and Higgins (1990) suggest that the average mass of males (517.6  $\pm$  64.32 g, 443–600 g, n = 3) is greater than that of females (423.5  $\pm$  9.40 g, 414–433 g, n = 2).

Wing length has been measured by Murphy and Pennoyer (1952) at 306.1  $\pm$  5.45 millimetres (296–317 mm, n = 25) for males and 302.1  $\pm$  7.59 millimetres (284–316 mm, n = 58) for females. The method used to sex these birds was not mentioned by the authors, although it was probably by dissection and internal examination. In Providence Petrels, the bill is laterally compressed and considerably hooked (Bailey *et al.* 1989). The culmen measures 34.6  $\pm$  1.05 millimetres (32.7–37.1 mm, n = 25) in males and 34.3  $\pm$  1.18 millimetres (30.1–36.2 mm, n = 58) in females (Murphy and Pennoyer 1952). Tarsi are rounded anteriorly with some slight compression, and have been measured at 42.6  $\pm$  1.09 millimetres (40.9–44.7 mm, n = 25) for males and 41.8  $\pm$  1.04 millimetres (39.1–44.6 mm, n = 58) for females (Murphy and

Pennoyer 1952). Despite males having slightly larger wings, culmen and tarsi, Murphy and Pennoyer (1952) found that the differences between sexes were not significant enough to conclude sexual dimorphism in this species.

The ability to sex individual Providence Petrels provides a means to investigate sex-specific differences in foraging and breeding behaviour. Unfortunately many bird species are very difficult to sex in the hand (Robertson et al. 2000), particularly seabirds. Based on the information available (Murphy and Pennoyer 1952; Marchant and Higgins 1990), it appears that male Providence Petrels are slightly heavier and have longer wings, culmen and tarsi. Some studies have used a discriminant analysis function to sex birds (Anderson 1975; Fullagar and Disney1981; Johnstone and Niven 1989), which is able to determine particular morphological characteristics that distinguish between sexes. Unfortunately this technique can be quite restrictive (Johnstone and Niven 1989) and is also subject to high error. Johnstone and Niven (1989) suggested that the inclusion of mass would restrict this procedure to the start of the incubation period, given changes in mass over the season. Nowadays most studies are using DNA analysis as a tool for sexing individuals. Molecular analysis is currently the most feasible and accurate (99.99%) method for determining sex (Griffiths et al. 1998). Another advantage of molecular analysis is that it can be conducted throughout the year.

Much morphological information regarding the Providence Petrel is still lacking and it is unclear whether this species is sexually dimorphic based on previous research. Small sample sizes provided by Marchant and Higgins (1990) and lack of information regarding the method used for sexing by Murphy and Pennoyer (1952) necessitate a more rigorous study to support their findings. This study will therefore use molecular analysis to accurately sex individuals and determine if this species is sexually dimorphic. Such information will be used in other studies of this species to determine any sex-specific differences in foraging and breeding behaviour. Lord Howe Island (31°33'S, 159°05'E) is located in the South Pacific Ocean, 770 kilometres northeast of Sydney and 800 kilometres northwest of New Zealand (Hutton 1991). Birds were studied on the summit of Mt Gower (Fig. 1).

In total, 36 adults were weighed and measured over a oneweek period during chick rearing in 2000. Adults were weighed to  $\pm$  5grams in calico bags using a 1000-gram Pesola spring balance. Weights were only taken after adults had finished feeding their chick. Culmen lengths and tarsus lengths were measured to  $\pm$  0.05 millimetres using dial callipers, and the maximum flattened chord (wing) of each bird was measured to  $\pm$  1.0 millimetre using a butt-stopped ruler. Adults were banded with individually numbered, stainless steel bands provided by the Australian Bird and Bat Banding Scheme.

Each bird was sexed by Dr. David Groth from molecular analysis of DNA taken from feather samples. Contour feathers were collected from the mantle using tweezers and stored in locktight plastic bags. Only feathers containing the node were kept. Feathers were sexed in the laboratory by extracting the DNA material from skin cells inside the feather shaft to detect the presence of a particular sex-linked gene (following Griffiths *et al.* 1998). This test gives an accuracy of 99.99% and is considered a better alternative than sexing adults from cloacal examination (Serventy 1956) or from vocalisation (Warham 1990). The mass and morphometric measurements of each DNAsexed bird were compared using paired *t*-tests (to avoid pseudoreplication of data, following Gray and Hamer 2001) to determine if, based on these particular morphological features, sexual dimorphism existed.

#### RESULTS

The DNA analysis confirmed the sex of 18 male/female breeding pairs. The mean mass of these males (507.2 ± 41.5 g, n = 18) and females (476.6 ± 44.7 g, n = 18) were found to be significantly different (Paired *t*-test: t17 = 3.6, P = 0.002). Similarly, culmen measurements were significantly different between males and females ( $36.5 \pm 1.1 \text{ mm}$  and  $35.5 \pm 1.3 \text{ mm}$ , respectively, n = 18; Paired *t*-test: t17 = 2.8, P = 0.012). No significant differences between sexes were found for tarsus measurements (Paired *t*-test: t17 = 1.1, P = 0.311), or wing chord (Paired t-test: t15 = 0.7, P = 0.526). Table 1 provides a comparison of measurements and weights taken from male and female birds.

### DISCUSSION

The mean mass of adult Providence Petrels in this study was 507 grams for males and 477 grams for females (n = 36). These mass figures are somewhat different to those in Marchant and Higgins (1990), although this may be a function of their smaller sample size (n = 5). Murphy and Pennoyer (1952) found slight



Figure 1. The location of the study area on Lord Howe Island.

## TABLE 1

Mass and lengths of culmen, tarsus and maximum flattened chord (MFC) of DNA sexed Providence Petrels in 2000

	Males	Females
Sample size	18	18
Mean mass (g)	507	477
SD (g)	41.5	44.7
Range (g)	420–640	405–625
Sample size	18	18
Mean culmen (mm)	36.5	35.5
SD (mm)	1.1	1.3
Range (mm)	34.9–38.1	32.9-37.8
Sample size	18	17
Mean tarsus (mm)	45.3	44.7
SD (mm)	1.1	1.3
Range (mm)	42.5–46.8	42.7–47.6
Sample size	17	16
Mean MFC (mm)	306	305
SD (mm)	5.8	6
Range (mm)	296–320	297-318

differences with wing length, culmen and tarsi between sexes, although results were not statistically significant. Results from this study support Murphy and Pennoyer's (1952) findings, with the inclusion of significant differences found with culmen length between sexes.

Thus, this study has shown that Providence Petrels are indeed sexually dimorphic based on greater mass and culmen lengths of males. Sexual dimorphism has only been recorded definitively for two other Pterodroma species: Trindade Petrel Pterodroma arminjoniana (Murphy and Pennoyer 1952), Grey-faced Petrel P. m. gouldi (Imber 1971; Johnstone and Niven 1989) and possibly in Gould's Petrel Pterodroma leucoptera leucoptera (Hindwood and Serventy 1941). All studies except for Hindwood and Serventy (1941) found that males were significantly heavier than females, while studies on the Grey-faced Petrel also recorded longer and deeper bills in male birds. Male Gould's Petrels were found to have longer culmen lengths (Hindwood and Serventy 1941), however, sample sizes were too small to positively support sexual dimorphism in this species (n = 5 males; n = 3 females). Johnstone and Niven (1989) used a discriminant function to sex Grey-faced Petrels with eight per cent error. A discriminant analysis function was also applied to the Providence Petrel, although it was subject to 11 per cent error based on greater mass and larger culmen length of males (Bester 2003). Thus, Providence Petrels should not be sexed in the hand using this technique as DNA analysis proves to be more reliable (only 0.01% error). Even though birds were found to be sexual dimorphic in this study it is suggested that other researchers should collect feathers and conduct DNA analysis to accurately sex individuals. DNA analysis can also be conducted at any time of the season, although there is lag time with the analysis.

The ability to sex individuals enables sex-specific differences in behaviour to be identified (Merton *et al.* 1984; González-Solís *et al.* 2000). This DNA analysis has been essential to other Providence Petrel studies investigating differences in foraging and breeding behaviour between sexes (Bester 2003).

#### ACKNOWLEDGMENTS

The Lord Howe Island Board provided logistical support for this study. Nicholas Carlile, Lisa O'Neill, Michael Jarman, Robert Wheeler and numerous other volunteers provided assistance in the field. Dr. David Groth undertook the molecular analysis of DNA extracted from feather samples. The maunscript was improved by comments from five anonymous referees.

#### REFERENCES

- Anderson, A. (1975). A method of sexing moorhens. Wildfowl 26: 77–82.
- Bailey, S. F., Pyle, P. and Spear, L.B. (1989). Dark Pterodroma petrels in the North Pacific: identification, status, and North American occurrence. American Birds 43: 400–415.
- Bester, A. J. (2003). 'The breeding, foraging ecology and conservation of the Providence Petrel Pterodroma solandri breeding on Lord Howe Island, Australia.' Ph.D. thesis, Charles Sturt University, Albury.
- Fullagar, P. J. and Disney, H. J. de S. (1981). Discriminant functions for sexing woodhens. Corella 5: 106–109.
- Fullagar, P. J., McKean, J. L. and Van Tets, G. F. (1974). Appendix F, report on the birds. In 'Environmental Survey of Lord Howe Island'. (Eds H. F. Recher and S. S. Clark) Pp. 55–72. (AGP: Sydney.)
- González-Solís, J., Croxall, J. P. and Wood, A.G. (2000). Sexual dimorphism and sexual segregation in foraging strategies of northern giant petrels, Macronectes halli, during incubation. Oikos 90: 390–398.
- Gray, C. M. and Hamer, K. C. (2001). Food-provisioning behaviour of male and female Manx shearwaters, Puffinus puffinus. Animal Behaviour 62: 117–121.
- Griffiths, R., Double, M. C., Orr, K. and Dawson, R. J. G. (1998). A DNA test to sex most birds. Molecular Ecology 7: 1071–1075.
- Hindwood, K. A. and Serventy, D. L. (1941). The Gould petrel of Cabbage Tree Island. Emu 41: 1–20.
- Hutton, I. (1991). 'Birds of Lord Howe Island: Past and Present.' (Ian Hutton: Coffs Harbour Plaza.)
- Imber, M. J. (1971). Filoplumes of petrels and shearwaters. New Zealand Journal of Marine and Freshwater Research 5: 396–403.
- Johnstone, R. M. and Niven, B. E. (1989). Sexing grey-faced petrels by discriminant analysis of measurements. Notornis 36: 261–265.
- Marchant, S. and Higgins, P. J. (1990). 'The Handbook of Australian, New Zealand and Antarctic Birds, Vol. 1.' (Oxford University Press: Melbourne.)
- Merton, D. V., Morris, R. B. and Atkinson, I. A. E. (1984). Lek behaviour in a parrot: the kakapo Strigops habroptilus of New Zealand. Ibis 126: 227-283.
- Murphy, R. C. and Pennoyer, J. M. (1952). Larger petrels of the genus Pterodroma. American Museum Novitates 1580: 1–43.
- Robertson, B. C., Millar, C. D., Minot, E. O., Merton, D. V. and Lambert, D. M. (2000). Sexing the critically endangered kakapo Strigops habroptilus. Emu 100: 336-339.
- Serventy, D. L. (1956). A method of sexing petrels in field observations. Emu 56: 213-214.
- Warham, J. (1990). 'The Petrels. Their Ecology and Breeding Systems.' (Academic Press: London.)