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The type of Rapa Shearwater *Puffinus (newelli?) myrtae* from the Austral Islands, Polynesia, with remarks on the morphological variation of the taxon

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SUMMARY.—Rapa Shearwater *Puffinus (newelli?) myrtae* is known solely from five specimens held in Tring and Paris museums, and from observations on the breeding islands. It has a potentially tiny breeding population that is apparently restricted currently to islets off Rapa (it formerly also bred on the main island of Rapa), as well as probably on Marotiri (south of Rapa), in the Austral Islands, Polynesia. Clear differences in morphology between the series in Paris and the holotype in Tring can be explained in terms of age-related variation, with the latter specimen being a juvenile. As a result, we describe adult plumage in detail for the first time. Genetic analysis confirmed that the Paris and Tring specimens can be unequivocally referred to the same taxon.

Bourne (1959) described *Puffinus assimilis myrtae* from a single female shearwater now held in the Natural History Museum, Tring, reported to have been collected on Rapa Island (27°37'S, 144°20'W; c.40 km²), in the south of the Austral archipelago (previously Tubuai Islands), Polynesia, in mid-April 1925, by H. J. Kelsall during the St. George Scientific Expedition (NHMUK 1925.12.22.147). Subsequently, four additional specimens were collected (now in Paris museum, see below), while a specimen, taken in May 1880 and held at the American Museum of Natural History, New York (AMNH), labelled as being from 'Marotiri' Island, actually belongs to the *P. assimilis* group, and was collected off New Zealand by Andreas Reischek, a well-known bird collector working in New Zealand (H. Shirihai pers. obs.; J.-C. Thibault pers. comm.). Austin *et al.* (2004) sequenced two of the Paris specimens and reported that *myrtae* was most closely related to Newell's Shearwater *P. newelli*, with the result that most subsequent commentators have considered *myrtae* a subspecies of *newelli*, with or without Townsend's Shearwater *P. auricularis* as a third race (e.g., Dickinson & Remsen 2013, del Hoyo & Collar 2014). Recently, however, it has been suggested that *myrtae* might most appropriately be treated specifically (Martínez-Gómez *et al.* 2015).

By inference, given his remark that 'it seems likely that a bird taken ashore in April was preparing to breed', Bourne presumably believed the Tring individual to be adult. Rapa and its associated islets have not been subject to frequent attention from ornithologists, but they were visited by the Whitney South Sea Expedition in April 1921 and February 1922, and more recently by J.-C. Thibault in October–December 1974 and A. Varney in December 1989–January 1990 (Thibault & Varney 1991). During the 1974 field work, four additional specimens of what has generally been assumed to be the same small black-and-white *Puffinus* were collected on Rapa's offshore islets and are now at the Muséum National d'Histoire Naturelle, Paris (MNHN 1975.1787–1790). However, recent detailed comparisons by HS of the Tring and Paris specimens revealed clear differences in plumage and biometrics between the type specimen of *myrtae* and the series in France. Here we describe the reasons

for this morphological variation, in the process describing adult *myrtae* for the first time, and provide genetic confirmation that the Tring and Paris specimens belong to the same taxon. Our study concludes that the type specimen is in fact a fledged juvenile, and not an adult as intimated by Bourne (1959).

Methods

Specimen analysis.—All relevant specimen material was examined by HS & VB, with the Tring *myrtae* being checked also by GMK; all measurements were taken by HS & VB according to standard protocols (Svensson 1992) using electronic callipers and a metal wing-rule with a perpendicular stop at zero: wing length (from carpal joint to tip applying gentle pressure to the primary-coverts—Svensson's method 2); tail length (from the pygostyle to the tip); tarsus length (from the back of the intertarsal joint to the last complete scute before the toes diverge) and bill length (from the tip of the maxilla to the feathers).

Molecular analysis.—A partial fragment of the mitochondrial gene cytochrome *b* (cyt *b*) of the holotype of *myrtae* was sequenced for comparison with sequences of other *Puffinus* species available on GenBank including those from two of the specimens collected on Rapa in Paris (MNHN 1975.1787, 1975.1788) (Austin *et al.* 2004). Total genomic DNA was isolated

TABLE 1
Conserved primer sequences used to amplify the mitochondrial cytochrome *b* gene; all those with the suffix *_neu* have been slightly modified from Pyle *et al.* (2011).

| Primer | Sequence |
|---------------------------|-----------------------------|
| aCytbPro3F | 5' CACACATGCCGAAAYGTACA |
| aCytbPro3R | 5' GCAGTTGCTATRAGRGRTRAG |
| aCytbPro4F _{neu} | 5' TAAAGAGACCTGAAACACAGG |
| aCytbPro4R | 5' CCCCTCAGGCYCATCTCTAC |
| aCytbPro5F _{neu} | 5' CAGCCATCCCATACATCG |
| aCytbPro5R _{neu} | 5' AATGGGATTTTTCACAGTTTG |
| aCytbPro6F _{neu} | 5' CACGAATCAGGCTCAAACAA |
| aCytbPro6R _{neu} | 5' TGGTTTGATATGAGGAGGTG |
| aCytbPro7F _{neu} | 5' CCTACTAGGAGAYCCAGAAAATTT |
| aCytbPro7R _{neu} | 5' GTTCGTTGTTTRGCCITGTG |

from a toe-pad sample of the *myrtae* holotype using sbeadex® forensic kit (LGC Genomics). Five fragments resulting in a partial sequence of cyt *b* were amplified with polymerase chain reaction (PCR) using slightly modified versions of the published primer pairs aCytbPro3F/aCytbPro3R, aCytbPro4F/aCytbPro4R, aCytbPro5F/aCytbPro5R, aCytbPro6F/aCytbPro6R and aCytbPro7F/aCytbPro7R (Pyle *et al.* 2011) (Table 1). PCR reaction volume was 25 µl containing 12.5µl GoTaq® Hot Start Green Master Mix (Promega), 2 µl genomic DNA, 2µl of each primer with a concentration of 10µM and 6.5

µl ddH₂O. PCR was performed using a SensoQuest Labcycler. The cycling conditions for amplification were: initial denaturation of 94°C for three minutes, followed by 40 cycles of denaturation at 95°C for 30 seconds, annealing for 30 seconds at 54°C and extension at 72°C for one minute, with a final extension at 72°C for seven minutes. PCR products were cleaned using the Wizard SV Gel and PCR Clean-UP System (Promega) or with Microsynth AG (Balgach, Switzerland). Sequencing was performed with Microsynth AG. Sequence preparation and editing of sequences was performed using Geneious Pro (Drummond *et al.* 2013).

Results

Molecular.—The resulting cyt *b* fragment from the *myrtae* holotype in Tring was 700 base pairs in length (GenBank accession no. KY933629) and differed in one synonymous substitution of a C with a T from the two sequences of *myrtae* from MNHN retrieved from GenBank (AY219938, 219939). We therefore eliminated the seemingly remote possibility that

TABLE 2

Mensural data taken by HS & VB for all known specimens of *Puffinus newelli myrtae*, measured according to standard parameters (see main text; Svensson 1992). For museum acronyms, see main text. Measurements of live birds (from Holyoak & Thibault 1984) are also given for comparison.

| Locality | Museum | Reg. no. | Date | Age | Sex | Wing | Tail | Culmen | Bill depth at hook | Bill width at gape | Tarsus | Mass |
|----------------------------------|--------|----------------|------------|----------|-----|--------|------|--------|--------------------|--------------------|--------|------|
| Rapa | MNHN | 1975.1787 | 23/10/1974 | Adult | F | 191.0 | 73.0 | 26.20 | 5.50 | 11.60 | 37.65 | |
| Rapa | MNHN | 1975.1789 | 20/10/1974 | Adult? | | 198.0 | 71.0 | 27.10 | 6.30 | 11.30 | 38.70 | |
| Rapa | MNHN | 1975.1788 | 20/10/1974 | Adult | F | 193.0 | 78.0 | 27.80 | 6.10 | 10.20 | 39.10 | |
| Rapa | MNHN | 1975.1790 | 16/10/1974 | Adult | F | 201.0 | 76.0 | 26.60 | 6.40 | 10.40 | 41.00 | |
| Rapa | NHMUK | 1925.12.22.147 | 17/04/1925 | Juvenile | F | 197.0 | 81.0 | 24.70 | 6.20 | 10.20 | 40.00 | |
| Means (excluding NHMUK specimen) | | | | | | 195.75 | 74.5 | 26.93 | 6.08 | 10.88 | 39.11 | |
| Mean for ten live birds | | | | Adult | - | 198.5 | - | 25.8 | - | - | 40.7 | 192 |

the Tring specimen was mislabelled as to locality or that it was a bird that had come ashore on Rapa from some other (perhaps even far) remote breeding grounds.

Morphology.—Our analysis of the five available specimens of *myrtae* (Table 2) revealed that the holotype in Tring is fairly distinct, being superficially most similar to the Southern Ocean Little Shearwater complex *P. elegans sensu lato* (although also very similar to larger forms assigned to *P. assimilis* in the New Zealand region) due to its distinctive greyish upperparts with extensive pale fringing, and especially the white vernal areas to the primaries. Also in agreement with Bourne (1959), we noticed its clearly proportionately long tail and very short bill. Below we demonstrate that the holotype is a different age (juvenile) to the Paris series (all of which are adults), describe the latter plumage for the first time and its differences from the first plumage.

The holotype is a juvenile.—As noted above, it appears that Bourne (1959) believed the Tring female to be an adult that had come ashore at the start of its breeding season. However, it is a juvenile, based on the following characteristics (Fig. 1), at least some of which are typical of juveniles of closely related taxa: short, narrow bill with an all-dark and smooth surface (lacking the scratches and well-developed bill plates, especially the hook and nasal tubes usual of adults); apparently fresh juvenile wing feathers, with more pointed primary tips; broadly white-fringed wing-coverts (as is often observed in juveniles of the closely related *P. assimilis* group); extensive pale grey fringes to the upperparts; and the weakly expressed breast-side patches.

Adult characters.—The following is principally based on an adult female (MNHN 1787.1975) collected on the offshore islet of Tauturou, on 23 October 1974. This and the other three Paris specimens all have adult-type feathers throughout, while their bills possess rather ragged and scratched surfaces, and brownish-coloured stains on their bellies, presumably due to soil, further indicates that they were breeding at the time. Note that specimen MNHN 1975.1789 is labelled as a fledgling, but this is a mistake as this individual has its feathers (e.g., upperwing-coverts) both worn and new, which excludes the possibility that it had recently fledged.

Upperparts blackish to greyish brown. Dark crown extends to upper edge of eye and is generally sharply demarcated, although at border with white ear-coverts there is a dark-mottled (streaked black to dull grey on white) superciliary region (= lores and behind eye to neck-sides). Some slate-tinged feathers are present on neck-sides (several of them tipped white) and scapulars. Basal sides of forehead also white but do not join above bill



Figure 1. Holotype of Rapa Shearwater, taxon *myrtae* (NHMUK 1925.12.22.147), a juvenile female, collected on Rapa Island, Polynesia, in April 1925. Note unique combination of greyish dorsal areas (1a) with white tips, especially to the greater wing-coverts (1b), and white undertail-coverts (1c), inner webs to the remiges (1c), and area above the eye (1d), which phenotypically place the taxon closer to the *P. assimilis* complex (see Relationships). Note also the narrow all-dark bill with smooth surface (1d), more pointed primary tips (1a), broadly white-fringed wing-coverts but weakly expressed breast-side patches (a & b) inferring a juvenile (Hadoram Shirihai, Tubenoses Project / © Natural History Museum, London)

because the dark crown reaches maxilla, with diffuse greyish-white upper forehead. Within the largely uniform head to tail and entire upperwing, the crown is noticeably blacker (appearing almost capped), as are the mantle, lesser coverts and carpal area to primaries. Clear demarcation between dusky upperparts and white underparts otherwise broken only by weak breast-side patches (dull blackish-brown mottling). Throat, breast and flanks



Figure 2. Dorsal, ventral and lateral views of two adult female Rapa Shearwaters, taxon *myrtae* (MNHN 1789.1975, below, and MNHN 1787.1975), collected on Tautourou, off Rapa Island, Polynesia, October 1974; unlike the holotype (a juvenile), note especially the much darker and more uniform upperparts (Hadoram Shirihai, Tubenoses Project / © Muséum National d'Histoire Naturelle, Paris)

clean white, partially separated from white undertail-coverts by blackish-brown divide formed by lateral extension of dark feathering extending narrowly from rump to thighs. Although undertail-coverts are predominantly white, some feathers (usually concealed) have inconspicuous, diffuse, greyish flecks and tiny patches. Underwing-coverts clean white, narrowly framed blackish brown on leading edge; remiges mostly dusky grey, with clear whitish basal inner webs to primaries, reaching to within *c.*3 cm of their tips; axillaries mainly white. No description of bare-parts coloration prior to collection. Now, bill appears mostly blackish with a slight slaty hue, while the inner tarsi, two inner toes and webs are paler than the otherwise blackish legs and feet.

Differences between juvenile and adults.—Bourne (1959) noted 'broad white feather edgings on the upper-parts' and we confirmed that the *myrtae* type is heavily scaled above (pale bluish-grey bases, subterminally darker and narrowly tipped whitish), but the four adults in Paris lack or virtually lack this feature, being far more uniformly dark in ground colour above. Furthermore, the juvenile *myrtae* type has obvious white fringes to the greater coverts (up to 2.5 mm wide), forming a clear wingbar, which is not seen in the four adults from Rapa. While pale / white tips can be lost with wear in these small shearwaters, the Paris specimens are still rather fresh, and overall much more uniformly black above than the *myrtae* type.

Bourne (1959) also reported that his type has 'a disproportionately-long tail', 81 mm (which measurement we confirmed), and both he and HS measured wing length very similarly, 196 mm (Bourne 1959) or 197 mm (right wing) and 198 mm (left wing), thus tail / wing ratio is 41.1. This is notably different from mean values for the four adults in Paris, which are 74.5 mm (tail), 196 mm (wing) and 38.0 (tail / wing ratio), respectively. Finally, Bourne (1959) and HS measured bill length as 25 mm and 24.2 mm, respectively, clearly indicating that the juvenile has a proportionately short bill (especially in relation to tail length, 29.9 tail / bill ratio). Mean bill length of the four adults in Paris = 26.9 mm (range 26.2–27.8 mm; tail / bill ratio 36.1). Additional measurements (of live birds) are available in Holyoak & Thibault (1984), including body mass (see Table 2).

Collection locality and conservation

The *myrtae* type was collected during the St. George Scientific Expedition. The expedition focused mostly on marine biology, with Kelsall the sole ornithologist on board, and no expedition log or publication on the birds collected on Rapa or the expedition as a whole is available. Thus, the precise location and circumstances of the type's collection are unknown. From the specimen labels and museum register, we may assume that it was collected on the main island of Rapa, as the collection date and the description of the expedition's visit in Collenette (1926: 236–261) correspond. Breeding by *myrtae* on the main island of Rapa is unknown today, with the only small shearwater ever claimed to have been collected on the main island being the *myrtae* type (Christmas Shearwater *P. nativitatis* nests in comparatively small numbers on some of the same offshore islets: Thibault & Varney 1991). However, bones have been attributed to this taxon from a rich archaeological excavation (Tennyson & Anderson 2012). Based on the observations by Thibault & Varney (1991), wherein it is listed under *P. assimilis*, *myrtae* (*c.*255–380 pairs) breeds only on small islets off Rapa, namely Tauturou, Rapa iti, Karapoo iti and Karapoo rahi, but these authors suggested that it might also nest around the cliffs at Haurei in the interior of the main island. Therefore, a survey of the main island to search for *myrtae* would be worthwhile (as already suggested by Thibault & Varney 1991).

Breeding of *myrtae* on Rapa seems seasonal, with birds arriving in April–May and departing in October (Holyoak & Thibault 1984); indeed, no birds were seen at breeding sites in December (Thibault & Varney 1991), March (C. Gaskin pers. comm., on Marotiri) or April (Beck ms, Quayle ms). Fledging time was clearly during October in 1975, with numerous large feathered chicks being found on Rapa’s offshore islets by J.-C. Thibault in late October, suggesting synchronous breeding (details in Holyoak & Thibault 1984; J.-C. Thibault pers. comm.). The *myrtae* type is a young bird, which was collected in April. Either it is a recent fledgling, raising the possibility of asynchronous breeding or twin breeding seasons (‘winter’ breeding on islets due to competition for burrows with Black-winged Petrel *Pterodroma nigripennis*, and ‘summer’ breeding on the main island or on Marotiri?), or it is a six-month-old juvenile that remained in the vicinity of Rapa, suggesting sedentary behaviour in this population. Visits to the colonies by adults in late November (Holyoak & Thibault 1984), c.1 month after fledglings had left, indeed suggests that post-breeding adults remain around Rapa.

Relationships

The most recent molecular hypothesis (Martínez-Gómez *et al.* 2015) for shearwaters in the relevant subclade of Austin *et al.* (2004) recommended that *P. auricularis* (which nests on Socorro Island, off north-west Mexico) and *P. newelli* (an endemic breeder to Hawaii) be treated as conspecifics, with the former name having priority for the single species, but that *myrtae* be separated at species rank. This hypothesis is based solely on genetic data (and only mitochondrial DNA), which indicate that *myrtae* is sister to a monophyletic clade comprising *auricularis* and *newelli*. Whereas the two last-named taxa differ by at most 0.6% in mtDNA (*cyt b*), *myrtae* differs by between 1.5% and 2.3% from both other taxa (Martínez-Gómez *et al.* 2015), although Pyle *et al.* (2011) reported that *newelli* and *myrtae* differ by just 1.2% in *cyt b*.

Morphological data provide support for the genetic hypothesis for *myrtae* of Martínez-Gómez *et al.* (2015). Rapa Shearwater is much smaller (in all biometric characters) than the other two taxa (see Table 3), and is furthermore highly distinctive in having a unique combination of white undertail-coverts (vs. dark in both *newelli* and *auricularis*) and whitish inner webs to the remiges, as well as a white face. All these features place it closer to the *Puffinus assimilis* complex (including *tunneyi*, *kermadecensis* and *haurakiensis* as subspecies) than to smaller forms previously assigned to Tropical Shearwater *P. lherminieri sensu lato*. Genetically, it is slightly closer to the recently described Bryan’s Shearwater *P. bryani* and to Black-vented Shearwater *P. opisthomelas* (Pyle *et al.* 2011) than to *P. assimilis* (3.8% and 2.6% vs. 4.2%), but *myrtae* differs from *bryani* in being larger and again in having whiter

TABLE 3

Mensural data taken by VB for three taxa of *Puffinus* shearwaters, measured according to standard parameters (see main text; Svensson 1992) and displaying mean values plus standard variation, and range.

| Taxon | Wing | Tail | Culmen | Bill depth at hook | Bill width at gape | Tarsus |
|--|-------------|------------|-------------|--------------------|--------------------|-------------|
| <i>Puffinus auricularis</i> (n = 20) | 228.5 ± 5.8 | 76.6 ± 2.6 | 31.09 ± 1.3 | 7.38 ± 0.6 | 11.99 ± 1.2 | 45.16 ± 1.1 |
| Range | 216–240 | 72–81 | 28.2–34.4 | 6.8–8.9 | 9.9–14.4 | 43.2–47.2 |
| <i>Puffinus newelli</i> (n = 60) | 233.6 ± 7.0 | 85.3 ± 4.4 | 33.24 ± 1.3 | 7.43 ± 0.5 | 13.02 ± 1.2 | 47.14 ± 1.4 |
| Range | 214–248 | 77–97 | 30.4–36.4 | 6.4–8.8 | 10.0–15.1 | 44.6–50.5 |
| <i>Puffinus newelli myrtae</i> (n = 5) | 195.8 ± 4.0 | 75.8 ± 4.0 | 26.52 ± 1.1 | 6.06 ± 0.4 | 10.74 ± 0.7 | 39.43 ± 1.4 |
| Range | 191–201 | 68.5–81 | 24.7–27.8 | 5.5–6.6 | 10.2–12.0 | 37.6–50.5 |

dark undertail-coverts and inner remiges, while *opisthomelas* is of similar size, but has quite different plumage. Genetic and morphological data available to date therefore suggest that *myrtae* might be best considered as a species on its own, pending further analyses, especially using other genes, especially nuclear markers.

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