

CYTOCHROME-B EVIDENCE FOR VALIDITY AND PHYLOGENETIC RELATIONSHIPS OF *PSEUDOBULWERIA* AND *BULWERIA* (PROCELLARIIDAE)

VINCENT BRETAGNOLLE,^{1,5} CAROLE ATTIE,² AND ERIC PASQUET^{3,4}

¹CEBC-CNRS, 79360 Beauvoir sur Niort, France;

²Villiers en Bois, 79360 Beauvoir sur Niort, France;

³Laboratoire de Zoologie Mammifères et Oiseaux, Museum National d'Histoire Naturelle, 55 rue Buffon, 75005 Paris, France; and

⁴Laboratoire de Systématique moléculaire, CNRS-GDR 1005, Museum National d'Histoire Naturelle, 43 rue Cuvier, 75005 Paris, France

ABSTRACT.—Although the genus *Pseudobulweria* was described in 1936 for the Fiji Petrel (*Ps. macgillivrayi*), its validity, phylogenetic relationships, and the number of constituent taxa it contains remain controversial. We tried to clarify these issues with 496 bp sequences from the mitochondrial cytochrome-*b* gene of 12 taxa representing three putative subspecies of *Pseudobulweria*, seven species in six other genera of the Procellariidae (fulmars, petrels, and shearwaters), and one species each from the Hydrobatidae (storm-petrels) and Pelecanoididae (diving-petrels). We also include published sequences for two other petrels (*Procellaria cinerea* and *Macronectes giganteus*) and use *Diomedea exulans* and *Pelecanus erythrorhynchos* as outgroups. Based on the pronounced sequence divergence (5 to 5.5%) and separate phylogenetic history from other genera that have been thought to be closely related to or have been synonymized with *Pseudobulweria*, we conclude that the genus is valid, and that the Mascarene Petrel (*Pseudobulweria aterrima*) and the Tahiti Petrel (*Ps. rostrata*) are distinct species. In trees constructed with maximum parsimony and maximum likelihood, *Pseudobulweria* is the sister taxon to *Puffinus* and *Calonectris*, and these genera in turn are most closely related to *Bulweria* (and *Procellaria* in the maximum-parsimony tree). *Pseudobulweria* is not closely related to *Pterodroma* in either tree. Because *Ps. r. trouessarti* from New Caledonia, and *Ps. r. rostrata* from Polynesia differ by only 0.6%, these taxa do not deserve species status and should be regarded as valid subspecies. Received 7 October 1996, accepted 23 July 1997.

THE GENUS *PSEUDOBULWERIA*, first proposed by Mathews in 1936 for the Fiji Petrel (*Ps. macgillivrayi*), remains one of the least-known genera of petrels (Warham 1996, Attié et al. 1997). Although it has been synonymized with *Pterodroma* (e.g. Jouanin and Mougouin 1979, Warham 1990, Del Hoyo et al. 1992), some systematists have reinstated it as a separate genus (Imber 1985, Warham 1996), either close to *Pterodroma* (Sibley and Monroe 1990) or to *Bulweria* and *Procellaria* (Imber 1985). In his reappraisal of the petrels, Imber (1985) also included in *Pseudobulweria* the Mascarene Petrel (*Ps. aterrima*), the Tahiti Petrel (*Ps. rostrata*), and a fossil taxon (*Ps. rupinarum*). Beck's Petrel (*Ps. rostrata becki*) is known from only two specimens and may be extinct. Similarly, *Ps. macgillivrayi* is known from only two specimens (Watling and Lewanavanua 1985), *Ps. aterrima* is known from seven specimens and is on the verge of extinction

(Attié et al. 1997), and *Ps. rupinarum* already is extinct (Olson 1975). Therefore, systematic studies on this group have relied nearly exclusively on *Ps. r. rostrata* and on morphological attributes assessed from museum specimens (e.g. Jouanin 1970, 1987; De Naurois and Erard 1979; Imber 1985).

Using available data as well as new information provided by DNA sequencing, we examined: (1) the validity of the taxon *aterrima* compared with *rostrata*, because these two taxa have been suggested to be conspecific (e.g. Jouanin 1970); (2) the validity of the genus *Pseudobulweria*; and (3) the phylogenetic positions of *Pseudobulweria* and *Bulweria* within the family Procellariidae. For this latter aspect, we paid particular attention to *Bulweria*, which has been claimed to be close to (Kuroda 1954) or synonymous with *Pterodroma* (Olson 1975), and together with *Pseudobulweria* is thought to be ancient (Bourne 1975; Imber 1985; Warham 1990, 1996). We used the mtDNA sequence (496 bp

⁵ E-mail: breta@cebc.cnrs.fr

segment) at the cytochrome-*b* locus because this segment has proved valuable for phylogenetic analysis at the species, generic, and familial levels in birds (e.g. Lanyon 1994, Baker et al. 1995, Helbig et al. 1995, Arctander et al. 1996, Friesen et al. 1996, Krajewski and King 1996).

MATERIAL AND METHODS

Species and sampling techniques.—We obtained samples from seven procellariid genera as well as one species of storm-petrel and one species of diving-petrel: Tahiti Petrel (*Ps. r. trouessarti*; New Caledonia, blood sample and *Ps. r. rostrata*; Gambier Is., blood), Mascarene Petrel (Réunion I., tissue), Barau's Petrel (*Pterodroma barau*; Réunion I., tissue), Black-winged Petrel (*Pt. nigripennis*; New Caledonia, blood), Bulwer's Petrel (*Bulweria bulwerii*; Canary Is., blood), Cory's Shearwater (*Calonectris diomedea*; Malta, blood), Wedge-tailed Shearwater (*Puffinus pacificus*; New Caledonia, blood), Northern Fulmar (*Fulmarus glacialis*; Brittany, tissue), Snow Petrel (*Pagodroma nivea*; Adélie Land, Antarctica, blood), Leach's Storm-Petrel (*Oceanodroma leucorhoa*; French Guyana, tissue), and Common Diving-Petrel (*Pelecanoides urinatrix*; New Zealand, tissue). We also used published sequences from *Procellaria cinerea* and *Macronectes giganteus* (Nunn et al. 1996), making available data from 9 of the 13 genera in this family (sensu Warham 1996). *Diomedea exulans* (Nunn et al. 1996) and *Pelecanus erythrorhynchos* (Avisé et al. 1994) were used as outgroup taxa. Blood samples (ca. 1 mL) were taken from the tarsus for the larger species and from the wing for the smaller species and preserved in a buffered solution of 4M guanidine-thiocyanate (see Laulier et al. 1995). Tissue samples (liver) were taken from specimens frozen at -18°C . One specimen was sampled for each species.

DNA extraction and PCR amplification.—A small amount of tissue or blood (ca. 0.1 g) was powdered in liquid nitrogen and suspended within CTAB buffer containing proteinase K (Doyle and Doyle 1987, Winpenninckx et al. 1993). Digestion was at 60°C for 1 h, and protein isolation was carried out with chloroform-isoamyl alcohol. In addition, 0.5 units of RNAase were added to the second aqueous phase and incubated at 37°C for 30 min to remove RNA. Total genomic DNA was precipitated with isopropanol. DNA concentration and quality were evaluated with a spectrophotometer. Extracted DNA was amplified with the following two primers, whose numbers correspond to the location of the 3' end of the primer respectively in the full sequence of human mtDNA (Anderson et al. 1981, in bold) and chicken mtDNA (Desjardins and Morais 1990): L14841/L14990 (5'-CA-TCCAACATCTCTGCTTGATGAAA-3') defined by Kocher et al. (1989), and H15338/H15487 (5'-GA-

TCCTGTTTCGTGGAGGAAGGT-3'). These primers amplify a DNA segment of 496 bases. PCR was performed in a 50- μL volume using ca. 0.3 μg of template DNA and 50 picomoles of each of the primers. The PCR mix (final concentrations) contained 20 mM Tris-HCL, pH 8.55, 16 mM $(\text{NH}_4)_2\text{SO}_4$, 2.5 mM MgCl_2 , 150 $\mu\text{g}/\text{mL}$ BSA, 330 μM dNTP, and 0.3 μL (1.5 units) of Goldstar *Taq* DNA polymerase (Eurogenetec). The PCR profile for amplifications was 35 cycles of 60 s at 93°C , 40 s at 50°C , and 40 s at 72°C . PCR products were opened under a specially designed hood and checked by electrophoresis in 1% agarose-BET and TBE buffer (Sambrook et al. 1989) with the molecular weight marker VI (Boehringer). PCR products were cloned using the PCRscript TM SK(+) cloning kit (Stratagene) according to recommended specifications. A classical white/blue selection (Sambrook et al. 1989) was used for screening recombinant clones. Four white colonies per cloning were grown overnight in L-broth at 37°C , the phagemidic DNA extracted (Sambrook et al. 1989), and the insert was checked by digestion of the recombinant phagemidic DNA with BssHII. Sequencing on microtiter plates was performed with the T7 sequencing kit (Pharmacia), using the method of terminator dideoxynucleotides (Sanger et al. 1977). Two colonies per cloning were sequenced with external vector primers KS and T3 to check for artifacts introduced by cloning of PCR products. Differences between clones occurred in only two samples (both one-transition difference in the 496 bp), and a third colony was sequenced to confirm the correct sequence.

Data analysis.—Sequences were read and entered twice using the computer package MUST (Philippe 1993) and were easily aligned because no insertions or deletions were found. Relative transitional saturation was examined by plotting transitional against transversal pairwise raw differences. Maximum-likelihood (ML) analyses (Felsenstein 1981) were performed with program FastDNAm1 (Olsen et al. 1994). The options F (empirical frequencies) and G (global rearrangements) were used, and 10 random-input order of species were done for each model to minimize input-order bias. We used three models differing in the number of categories of substitution rates (CSR). In 1-CSR, the three codon positions had equal substitution rates; in the 2-CSR model, first and second codon positions had equal rates whereas the third differed; and in the 3-CSR model, each codon position had a different rate of substitution. Two sets of relative codon-position evolutionary rates (1st:2nd for the 2-CSR model, and 1st:2nd:3rd for the 3-CSR model) were used: 1:5 and 1:10, and 2:1:5 and 2:1:10, respectively. Three transition-transversion ratios (TS:TV) were used and thus defined *a priori* in the likelihood computations: 2:1, 5:1, and 10:1. The best likelihood values were obtained for 2-CSR and 3-CSR models with TS:TV=5:1, and with evolution-

ary category rates being respectively 1:10 or 2:1:10. These parameters produced two trees that differed in the position of *Procellaria cinerea*, with either a basal position to the clade *Pterodroma-Fulmarus-Pagodroma-Macronectes* (Model 2-CSR), or an inner position in this clade, as sister-group of fulmarines (Model 3-CSR). However, because three short branches did not significantly differ from zero, these two trees effectively gave the same topology, and thus we have presented only this tree beyond.

We also performed maximum-parsimony (MP) analyses with PAUP 3.1.1 (Swofford 1991) with unordered characters. Branch and bound searches were performed with the complete data set of 14 taxa. Weights were given to the transversions via a step matrix. The two optimizations used, Accelerated Transformation (ACCTRAN) and Decelerated Transformation (DELTRAN), yielded similar topological results. Likelihoods of the MP trees were calculated and compared with those of ML trees. Bootstrapping (Felsenstein 1985) was performed using FastDNAmI and PAUP. For the MP trees, heuristic searches were used for 100 and 1,000 iterations. *Pelecanus erythrorhynchos* was used as an outgroup in all analyses.

RESULTS

Sequence variation.—Sequences were deposited in GenBank under accession numbers U70482 to U70493. Within the 14 species analyzed, 174 sites (35% of the 496 sites) were found to be variable, and 126 (25.4%) were phylogenetically informative, i.e. were present in two or more states in more than one species. Of the 174 variables sites, 32 (18%) were at the first position of the codon, seven (4%) at the second, and the remaining 135 (75.6%) at the third position, representing at each position 19.4, 4, and 81%, respectively, of all such sites. These are similar proportions to those found for albatrosses (Nunn et al. 1996) and are typical proportions in cytochrome-*b* gene of vertebrates (see Cicero and Johnson 1995, Arctander et al. 1996).

Sequence divergence.—Among taxa within the same genus, uncorrected sequence divergences ranged from 0.6% between the two subspecies of the Tahiti Petrel to 7.1% between the two species of *Pterodroma* (Table 1), values that are within the range found for other nonpasserine genera (Cicero and Johnson 1995, Helbig et al. 1995, Nunn et al. 1996). The sequence of the Mascarene Petrel differed from that of the two subspecies of the Tahiti Petrel by an average of 5.3%, typical of levels of divergence among

TABLE 1. Pairwise comparisons of the partial cytochrome-*b* sequence data (496 nucleotides); % global substitutions are below the diagonal, and % transversions in the variable sites are above the diagonal.

Taxon	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
1 <i>Pelecanus erythrorhynchos</i>	—	35.1	48.7	36.2	46.1	39.1	27.5	26.7	29.2	33.8	35.0	37.1	37.3	35.6	33.9	33.3
2 <i>Diomedea exulans</i>	15.1	—	37.3	34.3	34.8	34.8	27.6	33.8	36.4	28.8	36.7	32.9	34.9	33.9	35.4	34.8
3 <i>Oceanodroma leucorhoa</i>	15.3	15.1	—	32.6	39.0	39.0	31.8	31.4	31.0	28.4	39.5	36.6	35.3	38.9	42.0	41.4
4 <i>Pelecanoides urinatrix</i>	13.9	14.1	17.3	—	29.0	29.5	22.1	20.6	17.4	22.4	23.7	25.4	28.6	24.1	26.3	25.8
5 <i>Pterodroma nigripennis</i>	12.7	13.9	16.5	13.9	—	5.7	21.1	18.0	21.5	21.3	23.3	25.4	24.6	25.9	26.3	25.0
6 <i>Pt. barau</i>	13.9	13.9	15.5	12.3	7.1	—	23.2	15.2	19.7	22.5	21.4	21.2	21.9	22.6	26.5	25.0
7 <i>Procellaria cinerea</i>	16.1	15.3	17.1	13.7	14.3	11.3	—	10.9	13.0	17.0	9.4	10.6	9.2	9.2	9.8	9.4
8 <i>Pagodroma nioea</i>	15.1	13.7	17.3	12.7	12.3	11.9	11.1	—	7.4	22.5	8.8	10.8	10.1	9.1	9.8	10.0
9 <i>Fulmarus glacialis</i>	14.5	13.3	16.9	13.9	13.1	12.3	10.9	8.1	—	13.5	12.5	12.7	13.5	11.3	12.5	12.3
10 <i>Macronectes giganteus</i>	14.3	13.3	16.3	11.7	12.3	9.9	11.9	9.7	7.5	—	17.0	18.0	16.7	14.8	18.9	18.5
11 <i>Bulweria bulwerii</i>	12.1	12.1	14.3	11.9	12.1	11.3	10.7	11.5	9.7	10.7	—	12.5	9.8	8.7	7.1	6.9
12 <i>Calonectris diomedea</i>	12.5	14.7	16.5	12.7	12.7	13.3	13.3	13.1	12.7	12.3	9.7	—	11.4	13.1	13.2	12.9
13 <i>Puffinus pacificus</i>	11.9	13.3	17.1	11.3	13.1	12.9	13.6	12.9	11.9	12.1	10.3	9.1	—	9.6	12.0	11.7
14 <i>Pseudobulweria aterrima</i>	11.9	13.1	14.5	11.7	10.9	10.7	10.9	11.1	10.7	12.3	9.3	9.3	10.7	—	11.9	10.6
15 <i>Ps. rostrata trouessarti</i>	11.9	13.1	13.9	11.5	11.5	9.9	12.3	12.3	11.3	10.7	8.5	10.7	10.3	5.0	—	0.0
16 <i>Ps. rostrata rostrata</i>	12.1	13.3	14.1	11.7	12.1	10.5	12.9	12.1	11.5	10.9	8.7	10.9	10.5	5.6	0.6	—

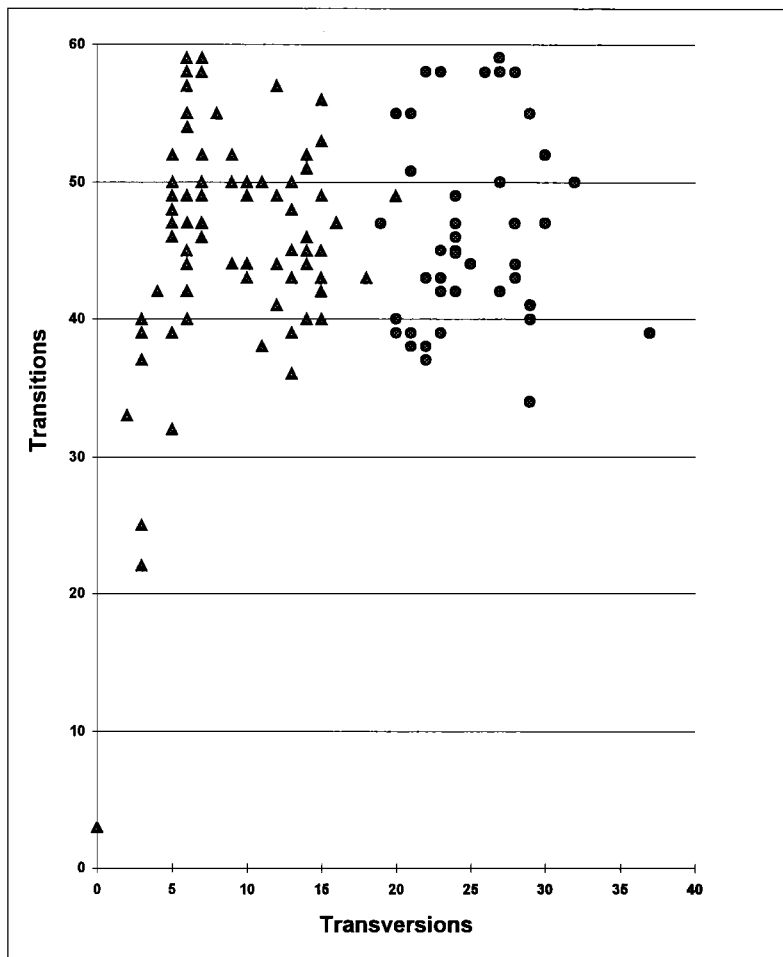


FIG. 1. Pairwise plots of transitions versus transversions (between Procellariidae, triangle; between the Procellariidae and other Procellariiformes or *Pelecanus*, circle).

congeneric species of birds. Among genera within the family Procellariidae, sequence divergences ranged from 8.1 to 14.3% (Table 1). Divergence between the Procellariidae sequenced in this study and other published sequences of procellariiforms ranged from 11.5 to 17.3%. Lastly, among the 14 taxa studied, the proportions of transversional substitutions in pairwise comparisons among taxa were highly variable, ranging from 0 to 0.49.

Saturation effects.—Relative saturation of transitions appeared fairly rapid (Fig. 1), which indicated that using transitions and transversions with equal weight for phylogenetic analysis was not acceptable. The very beginning of the curve, corresponding to the most closely related taxa, indicated that TS:TV ratio

was near 10:1 (Fig. 1; see also Austin 1996, Nunn et al. 1996), whereas the unweighted MP analysis indicated that the relative frequencies of TS:TV was 5:1. Therefore, we used both of these ratios for subsequent construction of MP trees.

Phylogenetic position of Pseudobulweria and Bulweria.—Although the topologies of the ML and MP trees (Figs. 2 and 3) differ somewhat in their placement of genera, in both trees *Pseudobulweria* is the sister group to *Puffinus* and *Calonectris*, and these genera in turn are most closely related to *Bulweria* (and *Procellaria* in the MP tree). *Pseudobulweria* is not closely related to *Pterodroma* in either tree. Bootstrap support is strong only for the monophyly of *Pseudobulweria* on the one hand and for the monophyly of

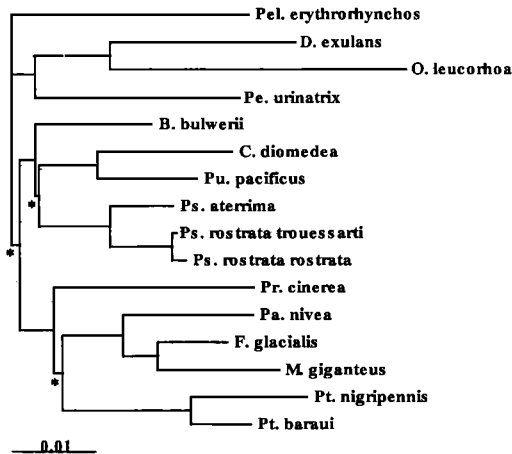


FIG. 2. Maximum-likelihood tree based on 496 bp of cytochrome-*b* DNA sequences with transition/transversion ratio (TS:TV = 5:1) and two categories of substitution rates (1:10; see Methods). The branches marked with a star are not significantly positive. Data for *Pelecanus erythrorhynchos* are from Avise et al. (1994); data for *Diomedea exulans*, *Macronectes giganteus*, and *Procellaria cinerea* are from Nunn et al. (1996).

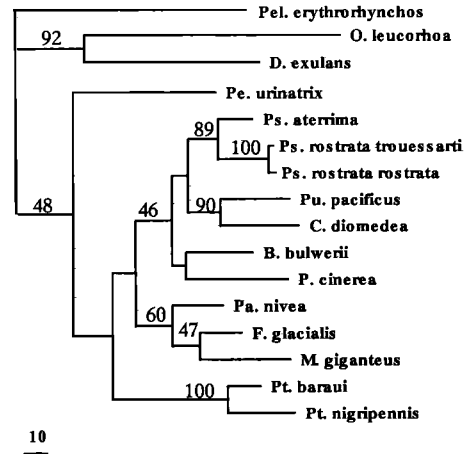


FIG. 3. Single most-parsimonious tree obtained using a TV:TS = 5:1 weighting scheme with bootstrap proportions (percentages) shown to the left of internal branches. The same topology was found with the 10:1 weighting scheme (see text for method and parameters). Bootstrap proportions are given only when >40%. Similar values were obtained with maximum-likelihood analysis.

Pterodroma on the other. Weaker support for other nodes is a common result when short cytochrome-*b* sequences are analyzed at this phylogenetic level (e.g. Wink 1995).

DISCUSSION

Validity and phylogenetic position of Bulweria and Pseudobulweria.—The genus *Pseudobulweria* has long been thought to be closely related to, or even included within, *Pterodroma*. Our genetic analyses do not support this belief; *Pseudobulweria* and *Pterodroma* are not members of the same clade within the Procellariidae (Figs. 2 and 3), and they are quite divergent. The same conclusion applies to *Bulweria*. Therefore, we conclude that *Pseudobulweria*, as suggested by Mathews (1936), is a valid genus, consisting of four to five species. Imber (1985) reached the same conclusion based on nongenetic evidence, and he further suggested that *Pseudobulweria* was linked to *Procellaria* and *Bulweria*, and more distantly, to *Puffinus*. In the ML tree (Fig. 2), *Pseudobulweria* and *Bulweria* were linked to *Puffinus* (and *Calonectris*) but apparently not to *Procellaria*, although the phylogenetic positions of *Bulweria* and *Procellaria* within the clade remain

imprecise due to low bootstrap values and/or sequences that are too short.

Subspecies of Pseudobulweria rostrata.—Three subspecies of the Tahiti Petrel currently are recognized: *rostrata*, *trouessarti*, and *becki* (Jouanin and Mougins 1979, Warham 1990). However, the validity of the first two subspecies is controversial (Murphy and Pennoyer 1952, De Naurois and Erard 1979), as is the taxonomic status of *becki*, which may require full species status (e.g. King 1978, Collar and Andrew 1988, Sibley and Monroe 1990), although all recent systematic studies of the Procellariiformes regard it as a subspecies (Jouanin and Mougins 1979, Imber 1985, Warham 1990). This latter taxon could not be included in our analysis because breeding colonies are undiscovered, but the other two subspecies were considered. Moreover, we sampled birds from the two geographic extremes: (1) New Caledonia, the westernmost breeding location of the species; and (2) the Gambier Islands, a newly discovered breeding site for this species (2,000 km east of its previous known range; Bretagnolle and Thibault unpubl. data). Our DNA sequence data show that populations from New Caledonia and Gambier are closely linked, differing by only three transitions and leading to the smallest percentage of difference among the

taxa we analyzed (0.6 %). This is at the lower end of sequence divergence found at the subspecific level among birds in general (see Helbig et al. 1995), and petrels in particular (e.g. Randi et al. 1989; Wink et al. 1993a, b), although Brooke and Rowe (1996) recently split a species of petrel with only 1% difference.

According to our molecular evidence, *trouesarti* from New Caledonia and *rostrata* from Polynesia do not deserve species status, and they should be regarded as valid subspecies. This conclusion is supported both by morphometrics and vocalizations (De Naurois and Erard 1979, Bretagnolle unpubl. data), although data on *rostrata* from Vanuatu and Fiji currently are lacking, and these birds may prove to be intermediate between the two forms.

Taxonomic status of Mascarene and Tahiti petrels.—Using external morphology, Jouanin (1970) suggested that the Mascarene and Tahiti petrels might be conspecific (along with *becki*). Our results from mtDNA show that the difference between the two petrels (5 to 5.5% sequence divergence; Table 1) involves 3 transversions and 20 to 22 transitions. This is clearly outside the range found between subspecies in birds, but within the range of divergence for congeneric species (Helbig et al. 1995). Bootstrap values also were high, and therefore it can be concluded that *Ps. aterrima* and *Ps. rostrata* are indeed valid congeneric species. Although nothing is known about the breeding ecology of *Ps. aterrima* (Attie et al. 1997), studies of external morphology have convincingly reached the same conclusion of species status (see Imber 1985).

ACKNOWLEDGMENTS

Field work was facilitated by J. Broudisou, M. Pandolfi, and S. Sirgouant on New Caledonia; by M. Attie on Réunion Island; and by J. R. King on the Canary Islands. WWF-France provided a grant to C.A. in 1995 for research on the endangered Mascarene Petrel. N. Hyde and J.-C. Stahl kindly provided tissue samples from The National Museum of Wellington, New Zealand, as well as F. Siorat (LPO, Station ornithologique de l'île Grande). Y. Bigot (Tours University) kindly allowed us to use his technique for blood preservation, and J.-F. Murail and P. Fondère helped with processing FastDNAm1 on UNIX computer at the Centre Informatique du Muséum. This work was conducted at, and supported by, the Service de Systématique Moléculaire (GDR 1005/CNRS, Muséum National d'Histoire Naturelle, Paris). Spe-

cial thanks to A. J. Baker for extensive revision of the manuscript, and to R. Dawson, S. J. G. Hall, A. Paterson, and two anonymous referees for improving a previous draft.

LITERATURE CITED

- ANDERSON, S., A. T. BANKIER, B. G. BARRELL, M. H. L. DE BRUIJN, A. R. COULSON, J. DROUIN, I. C. EPERON, D. P. NIERLICH, B. A. ROE, F. SANGER, P. H. SCHREIER, A. J. H. SMITH, R. STADEN, AND I. YOUNG. 1981. Sequence and organization of the human mitochondrial genome. *Nature* 290:457-465.
- ARCTANDER, P., O. FOLMER, AND J. FJELDSA. 1996. The phylogenetic relationships of Berthelot's Pipits *Anthus berthelothii* illustrated by DNA sequence data, with remarks on the genetic distance between Rock and Water pipits *Anthus spinoletta*. *Ibis* 138:400-415.
- ATTIÉ, C., J.-C. STAHL, AND V. BRETAGNOLLE. 1997. New data on the endangered Mascarene Petrel *Pseudobulweria aterrima*: A third 20th century specimen and distribution. *Colonial Waterbirds* 20: in press.
- AUSTIN, J. 1996. Molecular phylogenetics of *Puffinus* shearwaters: Preliminary evidence from mitochondrial cytochrome *b* gene sequences. *Molecular Phylogenetics and Evolution* 6:77-88.
- AVISE, J. C., W. S. NELSON, AND C. G. SIBLEY. 1994. DNA sequence support for a close phylogenetic relationship between some storks and New World vultures. *Proceedings of the National Academy of Sciences USA* 91:5173-5177.
- BAKER, A. J., C. H. DAUGHERTY, R. COLBOURNES, AND J. L. MCCLENNAN. 1995. Flightless Brown Kiwis of New Zealand possess extremely subdivided population structure and cryptic species like small mammals. *Proceedings of the National Academy of Sciences USA* 92:8254-8258.
- BOURNE, W. R. P. 1975. The lacrymal bone in the genus *Bulweria*. *Ibis* 117:535.
- BROOKE, M. DE L., AND G. ROWE. 1996. Behavioural and molecular evidence for specific status of light and dark morphs of the Herald Petrel *Pterodroma heraldica*. *Ibis* 138:420-432.
- CICERO, C., AND N. K. JOHNSON. 1995. Speciation in Sapsuckers (*Sphyrapicus*): III. Mitochondrial-DNA sequence divergence at the cytochrome-*b* locus. *Auk* 112:547-563.
- COLLAR, N. J., AND P. ANDREW. 1988. *Birds to watch*. ICBP Technical Publication No. 8. Cambridge, United Kingdom.
- DEL HOYO, J., A. ELLIOTT, AND J. SARGATAL (Eds.). 1992. *Handbook of the birds of the world*, vol. 1. Lynx Edicions, Barcelona.
- DE NAUROIS, R., AND C. ERARD. 1979. L'identité subsécificque des populations néocalédoniennes de

- Pterodroma rostrata* Peale 1848. Oiseau et la Revue Française d'Ornithologie 49:235-239.
- DESJARDINS, P., AND R. MORAIS. 1990. Sequence and gene organization of the chicken mitochondrial genome. *Journal of Molecular Biology* 212:599-634.
- DOYLE, J. J., AND J. L. DOYLE. 1987. A rapid DNA isolation procedure for small amounts of fresh leaf tissue. *Phytochemical Bulletin* 19:11-15.
- FELSENSTEIN, J. 1981. Evolutionary trees from DNA sequences: A maximum likelihood approach. *Journal of Molecular Evolution* 17:368-376.
- FELSENSTEIN, J. 1985. Confidence limits on phylogenies: An approach using the bootstrap. *Evolution* 39:783-791.
- FRIESEN, V. L., A. J. BAKER, AND J. F. PIATT. 1996. Phylogenetic relationships within the Alcidae (Charadriiformes: Aves) inferred from total molecular evidence. *Molecular Biology and Evolution* 13:359-367.
- HELBIG, A. J., I. SEIBOLD, J. MARTENS, AND M. WINK. 1995. Genetic differentiation and phylogenetic relationships of Bonelli's Warbler *Phylloscopus bonelli* and Green Warbler *P. nitidus*. *Journal of Avian Biology* 26:139-153.
- IMBER, M. J. 1985. Origins, phylogeny and taxonomy of the gadfly petrels *Pterodroma* spp. *Ibis* 127:197-229.
- JOUANIN, C. 1970. Le Pétrel noir de Bourbon. Oiseau et la Revue Française d'Ornithologie 40:48-68.
- JOUANIN, C. 1987. Notes on the nesting of Procellariiformes in Réunion. Pages 359-363 in *Studies of Mascarene Island birds* (A.W. Diamond, Ed). British Ornithologists' Union University Press, Cambridge.
- JOUANIN, C., AND J.-L. MOUGIN. 1979. Order Procellariiformes. Pages 48-121 in *Checklist of birds of the world*, vol. 1, 2nd ed. (E. Mayr and G. W. Cottrell, Eds.). Museum of Comparative Zoology, Cambridge, Massachusetts.
- KING, W. B. 1978. Endangered birds of the world: The ICBP bird Red Data Book, vol. 2, Aves, 2nd ed. IUCN, Morges, Switzerland.
- KOCHER, T. D., W. K. THOMAS, A. MEYER, S. V. EDWARDS, S. PÄÄBO, F. X. VILLABLANCA, AND A. C. WILSON. 1989. Dynamics of mitochondrial DNA evolution in animals: Amplification and sequencing with conserved primers. *Proceedings of the National Academy of Sciences USA* 86:6196-6200.
- KRAJEWSKI, C., AND D. G. KING. 1996. Molecular divergence and phylogeny: Rates and pattern of cytochrome *b* evolution in cranes. *Molecular Biology and Evolution* 13:21-30.
- KURODA, N. 1954. On the classification and phylogeny of the order Tubinares, particularly the shearwaters (*Puffinus*). Published by the author, Tokyo.
- LANYON, S. M. 1994. Polyphyly of the blackbird genus *Agelaius* and the importance of assumptions of monophyly in comparative studies. *Evolution* 48:679-693.
- LAULIER, M., E. PRADIER, Y. BIGOT, AND G. PÉRIQUET. 1995. An easy method for preserving nucleic acids in field samples for later molecular and genetic studies without refrigerating. *Journal of Evolutionary Biology* 8:657-663.
- MATHEWS, G. M. 1936. A note on the Black Fiji Petrel. *Ibis* 6:309.
- MURPHY, R. C., AND J. M. PENNOYER. 1952. Larger petrels of the genus *Pterodroma*. *American Museum Novitates* 1580:1-43.
- NUNN, G. B., J. COOPER, P. JOUVENTIN, C. J. R. ROBERTSON, AND G. G. ROBERTSON. 1996. Evolutionary relationships among extant albatrosses (Procellariiformes: Diomedidae) established from complete cytochrome-*b* gene sequences. *Auk* 113:784-801.
- OLSEN, G. J., H. MATSUDA, R. HAGSTROM, AND R. OVERBEEK. 1994. FastDNA Aml: A tool for construction of phylogenetic trees of DNA sequences using maximum likelihood. *Computer Applications in Biosciences* 10:41-48.
- OLSON, S. L. 1975. Remarks on the generic characters of *Bulweria*. *Ibis* 117:111-113.
- PHILIPPE, H. 1993. MUST: A computer package of management utilities for sequences and trees. *Nucleic Acid Research* 21:5264-5272.
- RANDI, E., F. SPINA, AND B. MASSA. 1989. Genetic variability in Cory's Shearwater (*Calonectris diomedea*). *Auk* 106:411-418.
- SAMBROOK, J., E. F. FRITSCH, AND T. MANIATIS. 1989. *Molecular cloning: A laboratory manual*, 2nd ed., vol. 3. Cold Spring Harbor Laboratory, Cold Spring Harbor, New York.
- SANGER, F., S. NICKLEN, AND A. R. COULSON. 1977. DNA sequencing with chain-terminating inhibitors. *Proceedings of the National Academy of Sciences USA* 74:5463-5467.
- SIBLEY, C. G., AND B. L. MONROE, JR. 1990. *Distribution and taxonomy of birds of the world*. Yale University Press, New Haven, Connecticut.
- SWOFFORD, D. L. 1991. PAUP: Phylogenetic analysis using parsimony, version 3.0. Illinois Natural History Survey, Urbana.
- WARHAM, J. 1990. *The petrels: Their ecology and breeding systems*. Academic Press, London.
- WARHAM, J. 1996. *The behaviour, population biology and physiology of the petrels*. Academic Press, London.
- WATLING, D., AND R. F. LEWANAVANUA. 1985. A note to record the continuing survival of the Fiji (MacGillivray's) Petrel *Pseudobulweria macgillivrayi*. *Ibis* 127:230-233.
- WINK, M. 1995. Phylogeny of Old and New World vultures (Aves: Accipitridae and Carthartidae) inferred from nucleotide sequences of the mi-

- tochondrial cytochrome *b* gene. *Zeitschrift für Naturforschung* 50c:868–882.
- WINK, M., P. HEIDRICH, U. KAHL, AND I. SWATSCHEK. 1993a. Inter- and intraspecific variation of the nucleotide sequence of the cytochrome *b* gene in Cory's Shearwater (*Calonectris diomedea*), Manx Shearwater (*Puffinus puffinus*) and the Fulmar (*Fulmarus glacialis*). *Zeitschrift für Naturforschung* 48c:504–509.
- WINK, M., P. HEIDRICH, AND D. RISTOW. 1993b. Genetic evidence for speciation of the Manx Shearwater *Puffinus puffinus* and Mediterranean Shearwater *Puffinus yelkouan*. *Vogelwelt* 114:226–232.
- WINNENPENNINCKX, B., T. BACKELJAU, AND R. DE WACHTER. 1993. Extraction of high molecular weight DNA from molluscs. *Trends in Genetics* 9:407.

Associate Editor: A. J. Baker