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Designation of a neotype for *Eudyptula minor* (Aves: Spheniscidae)

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European naturalists first encountered undoubted Little Penguins (Eudyptula minor (J.R. Forster, 1781)) in Dusky Sound, Fiordland, New Zealand, in March-April 1773 (Fleming 1982; Hoare 1982, p. 244; Andrews 1987). The type description for Aptenodytes minor did not identify a type specimen (see Medway 1976a & b). As Forster (1781) mentioned both Dusky Sound and Queen Charlotte Sound, Marlborough, in the type description ("dum enim in portu obscuro (Dusky Bay)...Deinde incolae Aesluarii Reginae Charlottae (Quen [sic] Charlotte's Sound)"), both sites are included in the type locality (Articles 73.2.3 & 76.1, ICZN 1999; Checklist Committee 2022). Queen Charlotte Sound is about 800 km northeast of Dusky Sound.

Two deeply divergent genetic lineages of Little Penguins have been identified from New Zealand: the Australian clade (E. m. novaehollandiae (Stephens, 1826)) in the south-east of the South Island (within 200 km of Fiordland), and the New Zealand clade (E. m. minor) throughout the country, including the Chatham Islands (Banks et al. 2002; Peucker et al. 2009; Grosser et al. 2015, 2016, 2017; Checklist Committee 2022). The two clades are sympatric in at least the Otago region, with limited interbreeding, and they are treated as full species by some authors (Grosser et al. 2015, 2016, 2017). Note that subspecies E. m. albosignata, chathamensis, iredalei, and variabilis are no longer recognised, and are treated as synonyms of E. m. minor (Checklist Committee 1990, 2010, 2022, contra Kinsky & Falla 1976, and Checklist Committee 1980).

The Australian clade of Little Penguin is considered to have colonised New Zealand between AD 1500 and 1900 (Grosser et al. 2015, 2016). The western margin of its distribution within New Zealand is poorly known (Grosser et al. 2015) and may have changed over time. We present evidence of an 'Australian' bird being found 275 km north of Dusky Sound in 2016 and note that Grosser et al. (2015) reported two 'Australian' birds at Westport, 590 km north of Dusky Sound. These two data points indicate that the zone of sympatry for the two clades may include all of Fiordland. It is therefore possible that Forster encountered birds of the Australian clade in Fiordland in 1773, whereas there is no evidence that birds of the Australian clade have ever occurred in the north of the South Island (including Queen Charlotte Sound).

There is currently no known way to separate individuals from the two clades of Little Penguins using external morphological characters. The two clades are separable based on mitochondrial DNA (minimum 30 mutations separating the clades at the control region HVRI; Banks et al. 2002; Grosser et al. 2015), microsatellite markers (19 loci) and at least 10 population-specific alleles of the seventh intron of the nuclear β-fibrinogen gene (β-fibint7) (Grosser et al. 2015), and most individuals can be separated by multivariate analysis of skeletal measurements (Grosser et al. 2017). While behavioural differences (including vocalisations) have been suggested between the two clades (Banks et al. 2002; Miyazaki et al. 2014; Grosser et al. 2015), the only reliable way to assign a living bird to either clade is using genetic methods (Banks et al. 2002; Grosser et al. 2015, 2017).

If a case were made for Forster having encountered and described birds of the Australian clade when in Dusky Sound, then E. minor would have priority over E. novaehollandiae. The next available name for the New Zealand clade is E. albosignata Finsch, 1874—a name that has previously been applied solely to the 'white-flippered' morphotype of Little Penguin, which has a distribution restricted to the Canterbury region, on the north-east coast of the South Island (Checklist Committee 1953, 1970; Kinsky & Falla 1976).

In order to pre-empt and prevent the confusion that would ensue if the taxon name E. minor was applied to the Australian clade of Little Penguin, we here designate a neotype for Eudyptula minor. The neotype is from Queen Charlotte Sound, and has been genotyped as being from the New Zealand clade of Eudyptula minor (Fig. 1). This designation will

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preserve nomenclatural stability for both New Zealand Little Penguin (*Eudyptula minor minor*) and Australian Little Penguin (*E. m. novaehollandiae*).

Neotypification

Under Article 75 of the Code, we designate the following specimen as the neotype for *Eudyptula minor* (Forster, 1781): NMNZ OR.030213, adult male, from Little Ngakuta Bay, Queen Charlotte Sound, Marlborough, New Zealand (41.2732°S 173.9682°E). Found dead (likely killed by a dog) on 20 November 2015, and collected by Daniel Palmer, Department of Conservation (DOC). The neotype is preserved as a study skin (Fig. 2), trunk skeleton, and tissue sample, in the ornithological collections of the Museum of New Zealand Te Papa Tongarewa. Its measurements are: bill length 36 mm, tarsometatarsus 32.5 mm, mid-toe + claw 48 mm, flipper 105 mm, tail 25 mm, weight 977 g, testes 17 x 6.5 mm (left) and 13 x 4 mm (right); external measurements as per Marchant & Higgins (1990).

Genotypification of the neotype for Eudyptula minor

DNA was extracted from either tongue muscle (from the neotype) or blood (10 samples from Taumaka/Open Bay Islands, September 2016, and one sample from Dusky Sound, November 2016; Fig 1C) using a DNeasy Blood and Tissue kit (Qiagen), following the manufacturer's instructions but eluting in a final volume of 60μ l of Buffer AE. The mitochondrial control region HVRI and β -fibint7 loci were PCR amplified and sequenced following Grosser *et al.* (2015). The DNA sequences have been deposited in the GenBank repository (accession numbers OP270794–OP270805 for HVRI and OP270806–OP270817 for β -fibint7).

Sequences contained either no indels (HVRI) or a single base pair indel (β -fibint7), and so were aligned manually. Haplotypes of the β -fibint7 locus were reconstructed with PHASE v2.1 in DNASP v6.12.03 (Librado & Rozas 2009). A median-joining network was constructed for each locus with PopART (Leigh & Bryant 2015).

The median-joining networks showed that the neotype specimen exhibited a New Zealand lineage HVRI haplotype and β -fibint7 alleles found predominantly in New Zealand lineage birds (Fig. 1A and 1B). Samples from Dusky Sound and Taumaka also mainly possessed New Zealand lineage HVRI haplotypes, with only a single individual from Taumaka found with the Australian lineage (Fig. 1A). This individual also had two β -fibint7 alleles found mainly in Australian lineage birds (Fig. 1B), with one other Taumaka bird also exhibiting one copy of this allele. The remaining Taumaka samples and the Dusky Sound bird had β -fibint7 alleles found predominantly in New Zealand lineage birds (Fig. 1B).

Qualifying conditions for neotypification

In compliance with the qualifying conditions required for neotypification by Article 75.3 of the Code, our action here:

- (1) expressly aims to clarify the nominal identity of *Eudyptula minor* (Forster, 1781) and its type locality, in a situation where two taxa are potentially involved (Article 75.3 introduction, and clause 75.3.1);
- (2) defines, in Figure 1 (and 'Genotypification of the neotype'), the genetic characters by which the taxon *E. minor* is circumscribed from *E. novaehollandiae*; the neotype has those traits, as specified in its designation above (Article 75.3.2);
- (3) provides, in the above designation, data and description sufficient to identify the specimen designated (Article 75.3.3);
- (4) gives reference to sources (Medway 1976a & b) which establish that no type material survives (Article 75.3.4);
- (5) chooses as neotype an adult matching Forster's (1781) original description from a Forster-cited population most likely to hold the "New Zealand" genotype (which it does) (Article 75.3.5);
- (6) selects the neotype from a site that is part of the original type locality (Article 75.3.6); and
- (7) records the deposition of the neotype in a recognised scientific research institution (Article 75.3.7).

We thank Daniel Palmer (DOC) for providing a Little Penguin specimen from Queen Charlotte Sound that met the criteria for neotypification, Timothée Poupart (Te Papa) for providing Little Penguin blood samples from Taumaka / Open Bay Islands, Jean-Claude Stahl (Te Papa) for images of the neotype, and Richard Schodde for helpful comments that improved this manuscript.

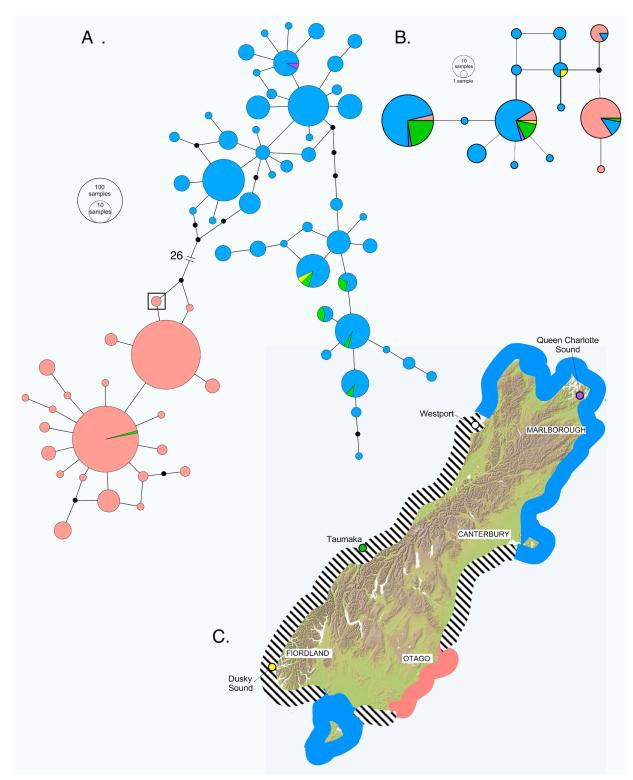


FIGURE 1. Median-joining networks of Little Penguin (A) HVR1 haplotypes and (B) β-fibint7 phased alleles based on data from Grosser *et al.* (2015) combined with newly generated sequences from this study. Intermediate unobserved haplotypes are shown as small black circles. Sequences from Grosser *et al.* (2015) are coloured by mtDNA lineage (blue = New Zealand lineage; pink = Australian lineage) and newly generated sequences are coloured by location, with the neotype shown in purple. Two of Grosser *et al.*'s (2015) HVR1 sequences from Westport exhibited an Australian lineage haplotype, which is indicated by a box. Circle size is proportional to haplotype frequency. (C) Map of the South Island distribution of Little Penguins, modified from Grosser *et al.* (2015). Ranges of the HVR1 lineages are indicated (blue = New Zealand lineage; pink = Australian lineage), with the grey dashed line showing the area where lineage distribution is uncertain. The sampling locations of newly generated sequences (Queen Charlotte Sound (neotype), Taumaka and Dusky Sound) are indicated.



5 cm

FIGURE 2. Little Penguin neotype (Museum of New Zealand Te Papa Tongarewa OR.030213, Little Ngakuta Bay, Queen Charlotte Sound, New Zealand), lateral, dorsal and ventral views. Images © Te Papa.

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