Running Head: What was the Canary Islands Oystercatcher?

Taxonomic status of the extinct Canary Islands Oystercatcher *Haematopus meadewaldoi*

TEREZA SENFELD,¹ THOMAS J. SHANNON,¹ HEIN VAN GROUW,² DANE M. PAIJMANS,³ ERIKA S. TAVARES,⁴ ALLAN J. BAKER,^{4§} ALEXANDER C. LEES^{5,6} & J. MARTIN COLLINSON¹*

¹School of Medicine, Medical Sciences and Nutrition, University of Aberdeen. Institute of Medical Sciences, Foresterhill, Aberdeen AB25 2ZD, UK

²Bird Group, Dept. of Life Sciences, The Natural History Museum, Akeman Street, Tring, Herts, HP23 6AP, UK

³FitzPatrick Institute of African Ornithology, DST-NRF Centre of Excellence, University of Cape Town, Rondebosch 7701, South Africa

⁴Department of Ornithology, Royal Ontario Museum, 100 Queen's Park M5S 2C6, Toronto, Canada

⁵School of Science and the Environment, Manchester Metropolitan University, Manchester, M1 5GD, UK

⁶Cornell Lab of Ornithology, Cornell University, Ithaca, NY 14850, USA

§ Deceased

*Corresponding author. Email: <u>m.collinson@abdn.ac.uk</u>

Mitochondrial genes were sequenced from four specimens of the extinct Canary Islands Oystercatcher *Haematopus meadewaldoi*, and compared to African Oystercatcher *H. moquini*, Eurasian Oystercatcher *H. ostralegus*, and an old unidentified extralimital 'black' oystercatcher specimen from The Gambia. At these loci, *H. meadewaldoi* was approximately 99.65% identical to multiple Eurasian Oystercatcher samples and in phylogenetic trees fell within the range of genetic variation observed in that species. The mystery Gambian bird was resolved as an extralimital *H. moquini*. We conclude that *H. meadewaldoi* was most likely a recently diverged melanistic morph or subspecies of *H. ostralegus*, though further genomic studies will be required to determine if there has been a period of isolation followed by introgression.

Keywords: Oystercatcher, Canary Islands, The Gambia, phylogeny, genetics

Oystercatchers (Aves: Haematopodidae) are a pan-global family of black or pied shorebirds for which, currently, eleven extant species are recognised (Gill & Donsker 2019). Reconstructing their phylogenetic relationships has been a longstanding challenge, due to their morphological, vocal and ecological similarities (Hayman et al. 1986). Even with the emergence of molecular data for some species, their relationships remain highly contentious because of shallow branching patterns and incomplete geographic sampling. Although nearly all species are either 'pied' or 'black', the Variable Oystercatcher Haematopus unicolor, a New Zealand endemic, has both black and pied morphs that overlap geographically and interbreed (Baker 1937), suggesting that plumage patterns are interchangeable and potentially not a good indicator of evolutionary relationships. Another New Zealand endemic, the South Island Oystercatcher H. finschi, was long considered a subspecies of the Eurasian Oystercatcher *H. ostralegus* which it closely resembles, but was given species status largely based on mitochondrial DNA (mtDNA) analyses (Banks & Paterson 2007). In North America, a black species, Black Oystercatcher *H. bachmani*, and a pied species, American Ovstercatcher *H. palliatus*, partially overlap in breeding ranges and are known to hybridise but have been classed as separate following a morphological analysis by Jehl (1985). More recently, there have been suggestions to lump them into a single species due to their mtDNA similarity (Herbert et al. 2004).

The taxonomic status of the poorly known Canary Islands Oystercatcher *H. meadewaldoi*, which became extinct sometime before 1940, has proven controversial. Only eight specimens exist: three at the Natural History Museum (Tring, UK, henceforth NHM), including the type specimen collected by Meade-Waldo in April 1888, a second collected in April 1890, and the last known specimen collected by Bannerman in April 1913. Elsewhere, two (both from Fuerteventura, 1889) are held at the Zoological Research Museum Alexander Koenig (Bonn, Germany); one (La Graciosa, 1890) is at Liverpool World Museum (UK) and two (Fuerteventura, 1889) at Manchester Museum (UK). The taxon was formally described by Bannerman as a geographically isolated subspecies of the black African Oystercatcher *H. moquini* of southern Africa, which it very closely resembles (Bannerman 1913). Subsequently it was treated as the only 'black' subspecies of the pied Eurasian Oystercatcher by Stresemann (1927) and Peters (1934), but again as subspecies of African Oystercatcher by Vaurie (1965) and Wolters (1975-1982). It was subsequently elevated to species status differing from African Oystercatcher on mean wing, bill and tarsal measurement (Hockey 1982). However, given its extinction, the range of morphological and plumage variation in the taxon will never be fully understood. The factors underpinning the disappearance of Canary Islands Oystercatcher are poorly known, but overharvesting of its invertebrate prey and holistic disturbance of the marine ecosystem have been implicated (Hockey 1987, Valledor de Lozoya 2013). It seems likely that predation by introduced cats and rats and potentially direct hunting by humans were also important (Collar & Stuart 1985). These threats are thought responsible for the global extinction of other sympatric coastal birds such as the Dune Shearwater *Puffinus holeae* (Rando & Alcover 2010).

The African Oystercatcher breeds no closer to the Canary Islands than Namibia (~4500 km), and it is highly philopatric (Bray & Hockey 2015). However, extralimital 'black' oystercatchers have been observed on several occasions further north on the African coast (Hayman *et al.* 1986). One such bird exists as a specimen at the NHM. According to its accession record, it is a female, captured alive in The Gambia during a 1938 expedition to West Africa by Jean Delacour and Lord Moyne and brought back to the UK where it was kept in an aviary until its death. Its bill grew abnormally long during confinement because of artificial feeding, and it is now considered impossible to identify the specimen biometrically, although Valledor de Lozoya (2013) tentatively suggested a fit with Canary Islands Oystercatcher. The possibility that it represented the last known Canary Islands Oystercatcher has not yet been excluded.

In order to investigate the taxonomic relationships of Canary Islands Oystercatcher and identify the Gambian specimen, we have sequenced mitochondrial genes from available specimens of these birds and also from African Oystercatchers, for which no sequences were publicly available.

METHODS

Toepads were sampled from the unidentified black oystercatcher at NHM Tring (; Supplementary Figure 1), from Canary Islands Oystercatcher specimens at the Liverpool World Museum (T.16000), Manchester Museum (B.9162; Supplementary Figure 2), and NHM Tring (NHMUK.1905.12.22.322 and NHMUK.1939.12.9.25). Feathers of a single Eurasian Oystercatcher were collected from a freshly dead bird in Scotland, UK, and blood samples of six African Oystercatchers were collected in Western Cape, South Africa. Ethical clearance (2011/V23/PH) was granted by the Science Faculty Animal Ethics Committee at The University of Cape Town. DNA was extracted using the QIAGEN QIAamp DNA Micro kit (for feathers and toepads) as per the manufacturer's instructions, with the addition of 0.1 M dithiothreitol to the proteinase K digest, and the QIAGEN QIAamp Blood Mini kit for blood samples. Details of individuals sampled are listed in Table 1.

Extensive precautions were taken to prevent and, if present, detect contamination. DNA was isolated from museum specimens separately from and prior to fresh material being handled. Separate reagents and plastics were used for DNA extractions and PCRs from museum and fresh material. All pipettes, plastic tubes and filter-tips were UV-crosslinked for several hours after each use. Potential contaminating DNA on pipettes was periodically denatured or depurinated with bleach or 0.25 M HCl. Canary Island Oystercatcher skin samples were processed and sequenced independently at two locations: those from NHM Tring by EST in Toronto in a dedicated ancient DNA facility, and those from Liverpool World and Manchester Museums by TS and TJS in clean conditions in Aberdeen. DNA from African Oystercatchers was isolated separately by DMP in Cape Town. PCRs were set up in clean sterile hoods. Water blanks were used as negative controls throughout, and PCRs and gels from museum and modern DNA were run separately. No incidences of contamination were observed at any time.

For Eurasian and African Oystercatchers the 5' "barcoding" region of *COI* and the entire *cytb* and *NADH2* genes were amplified in single polymerase chain reaction (PCR) runs with universal bird primers. Primers BirdF1/BirdR1 were used for *COI*, L14993/H16065 for *cytb*, and L5216/H6313 for *NADH2* with PCR reactions and conditions as previously described (Helbig & Seibold 1999, Herbert *et al.* 2004, Shannon *et al.* 2014).

Due to degradation of DNA in museum specimens, custom primers for Canary Island Oystercatcher and the mystery oystercatcher were designed based on consensus sequence obtained from Eurasian and African Oystercatchers. These primer pairs each amplified overlapping regions of up to ~220 bp such that the desired gene could then be assembled as a contig (Table S1). In the case of the mystery oystercatcher specimen, DNA could only be amplified in 80-100 bp fragments.

PCR products were run on a 1.5% agarose gel in 1x TAE buffer and ethidium bromide, and extracted using the QIAGEN Gel Extraction Kit. DNA concentrations were measured using a Nanodrop spectrophotometer and diluted to $10\mu g/\mu L$ for Sanger sequencing by Source Bioscience (Livingston, UK) with appropriate primers. Genbank accession numbers are listed in Table 1. No fragments > 250 bp could be obtained from any museum specimen, confirming absence of modern DNA contamination.

Alignments of assembled sequences were initially performed in CLC Sequence Viewer 8 (https://www.qiagenbioinformatics.com/products/clc-sequence-viewer/). To eliminate the possibility of pseudogenes or other nuclear copies, reading frames were verified by translation and sequences were compared with the same genes from all other available *Haematopus* species downloaded from NCBI Genbank (https://www.ncbi.nlm.nih.gov/nucleotide/) to confirm that in all cases the correct mitochondrial alleles had been isolated. Pied Avocet *Recurvirostra avosetta* and Black-winged Stilt *Himantopus himantopus* were used as outgroup taxa for tree rooting.

All sequences were aligned with default parameters in CLC Sequence Viewer. Alignments were trimmed by eye and positions with ambiguous bases due to poor sequencing reads were manually removed. A concatenated alignment of the three genes was generated to assess sequence

divergence between *H. ostralegus*, *H. moquini* and *H. meadewaldoi*. An unequal alignment was generated to include all available data from all published and new *Haematopus* sequences for generating a maximum-likelihood tree.

Maximum-likelihood trees were generated to inform the indicative mitochondrial relationships of oystercatcher species using W-IQ-TREE (Nguyen *et al.* 2015). The best-fit substitution model, TPM3+F+I, was selected using Model Finder (Kalyaanamoorthy *et al.* 2017). Bootstrap values based on 1000 pseudoreplicates were estimated using UFBoot2 (Hoang *et al.* 2017).

RESULTS

In order to resolve the taxonomic status of the Canary Islands Oystercatcher *H. meadewaldoi* and to identify the mystery oystercatcher captured in The Gambia in 1938, DNA was isolated from the specimens and three mitochondrial genes were amplified by PCR and sequenced.

A 2835 base pair (bp) concatenated sequence of *COI*, *cytb* and *NADH2* was generated for one Eurasian Oystercatcher and four African Oystercatchers; 1710 bp was obtained for two Canary Islands Oystercatchers (birds from Liverpool and Manchester Museums); 714 bp of *COI* sequence was obtained from the two NHM specimens. Across these three genes, Eurasian Oystercatcher differs from African Oystercatcher by an uncorrected-p distance of 0.63% (18 bp). This suggests a relatively short period of divergence, probably less than 150,000 years (Lavinia *et al.* 2016, Weir &Schluter 2008). Partial *COI* sequences of all four Canary Islands Oystercatcher were identical. The 1710 bp partial *COI/cytb/NADH2* Canary Islands Oystercatcher sequences were 2 bp different from each other, differing from the Eurasian Oystercatcher concatenation by only 6 bp (0.35%) and from African Oystercatcher by 19 bp (1.1%).

All published mtDNA sequences of multiple individuals of Eurasian and other species of oystercatcher were downloaded and aligned with new sequences obtained in this study. Data are unavailable only for the primarily Australian Pied *H. longirostris* and Sooty Oystercatchers *H*.

fuliginosus. A maximum likelihood tree drawn on the basis of the alignment (Figs. 1 and S3) recovered a strongly supported clade (98% bootstrap support) containing Canary Island Oystercatcher and Eurasian Oystercatchers of the nominate (European) and *H. o. longipes* (Russian/Caspian/Aral) subspecies to the exclusion of African Oystercatchers and all other oystercatcher taxa, including *H. ostralegus osculans* of Kamchatka and Korea, which may itself be a candidate for full species status. On the basis of mitochondrial data, Canary Islands Oystercatcher falls robustly within the range of genetic variation in Eurasian Oystercatcher

The quantity and quality of DNA obtained from the mystery oystercatcher from The Gambia were extremely poor, most likely due to post-mortem degradation. However, 281 bp of mitochondrial sequence (partial *COI* and *cytb* sequence) were eventually obtained spanning four bases that diagnostically separate all individuals of African Oystercatcher from Canary Island Oystercatcher. At each site the Gambian bird was identical to African Oystercatcher (Figs. 2, S4 and S5). Biometric data were taken from the Gambian bird but were inconclusive – the bill is overgrown due to its extended period in captivity (culmen 89.3 mm), outer primary heavily abraded to about half its length, and second primary only half-grown. However, the tarsus length (58.7 mm) is more consistent with female African Oystercatcher (mean 57.7 mm, range 52.0-62.0 mm, n = 54) than Canary Islands Oystercatcher (mean 53.9 mm, range 51.6-55.0 mm, n = 4) (Valledor de Lozoya 2013).

DISCUSSION

We found the Canary Islands Oystercatcher to be genetically more closely related to the Eurasian Oystercatcher than to the African Oystercatcher, despite its phenotypic similarity to the latter. Although no sequence data were previously publicly available for Canary Islands or African Oystercatchers, several of the specimens of Canary Islands Oystercatcher have been destructively sampled by other groups, and our genetic conclusions are in concordance with an unreviewed preliminary *COI* study in Valledor de Lozoya (2013). Using short sequence fragments at diagnostic loci, we were able to suggest that the 'mystery' Gambian oystercatcher specimen was a vagrant African Oystercatcher, at least 4500 km outside of its range. The hypothesis that it was the last recorded Canary Islands Oystercatcher was therefore rejected. Assuming that its appearance in the region was natural and not following human assistance, it ought to constitute the first record of African Oystercatcher for The Gambia.

Extensive precautions were taken to avoid the contamination of museum specimen samples with modern DNA (detailed in methods). We believe that no such contamination occurred and that the sequences reported for Canary Islands Oystercatchers and the Gambian bird are robust: Canary Island Oystercatcher DNA was isolated and sequenced in separate labs in Canada and UK, several years apart, gave identical results, and although Canary Island Oystercatcher sequences were similar to those of Eurasian Oystercatchers processed in our labs, they were not identical, eliminating the possibility of contamination. That no fragments larger than 220 bp could be amplified from museum DNA (and in the case of the Gambian bird no fragment larger than 100 bp) confirmed there was no contaminating modern oystercatcher DNA in any of those samples.

A close genetic relationship between Canary Islands and Eurasian Oystercatchers, to the exclusion of African Oystercatchers, confirms early suspicions based on morphometric data that African and Canary Islands Oystercatchers are distinct. The shorter bill length of African Oystercatcher is sexually dimorphic, a feature common to 'black' oystercatchers (Hockey 1982). Canary Islands Oystercatcher in this respect is more similar to 'pied' oystercatchers which tend to have longer bills and less variation between the sexes (Hockey 1982).

Although we cannot yet rule out the possibility that a very close, but reciprocally monophyletic relationship exists between Eurasian and Canary Island Oystercatchers, the data suggest that the Canary Islands Oystercatcher may represent a local melanistic subspecies of Eurasian Oystercatcher at the edge of its range. Island melanism is not uncommon in birds, especially on small islands (Uy & Vargas-Castro 2015, van Grouw 2017), and can occur with very little underlying genetic change: for example, in the Bananaquit *Coereba flaveola* melanistic

morphs in island populations are associated with mutation in a single pigmentation gene, *MC1R* (Theron *et al.* 2001).

Among the several unresolved contentious taxa of oystercatchers is the 'Far Eastern' Oystercatcher *H. ostralegus osculans*. While currently not recognised as a species, several studies have pointed out that its status should be reevaluated. A conservation assessment of *H. ostralegus osculans* notes that its longer bill, distinct juvenile and non-breeding plumage, and geographic isolation suggest that it should be considered an independent evolutionary unit (Melville *et al.* 2014). Previously, a morphological study of shorebirds classed *osculans* as a separate species (Livezey 2010) which would be consistent with our mitochondrial molecular phylogeny.

This study was; however, not intended to robustly resolve a full oystercatcher phylogeny, but to compare Canary Islands Oystercatcher with new and previously published sequence data. There is the possibility that Canary Islands Oystercatcher experienced a longer period of divergence than suggested here, but has subsequently experienced mitochondrial introgression from Eurasian Oystercatcher, which is an irregular visitor to the Canary Islands. In the future, a full genomic study would resolve this possibility, however, on the basis of current data we conclude that Canary Islands Oystercatcher was very closely allied to Eurasian Oystercatcher, with which it is possibly best considered conspecific.

TS performed this work as part of her MSc (Genetics) at University of Aberdeen. TJS is supported by PhD funding from The Sound Approach. DMP's research forms part of the Oystercatcher Conservation Programme and was supported by the National Research Foundation and the University of Cape Town. We thank curatorial staff, Tony Parker at the World Museum, Liverpool and Henry McGhie at Manchester Museum. We thank Amy Bastow for securing material from Eurasian Oystercatcher and Lucie Goodayle for photography of NHM oystercatcher specimen. EST was funded by the Canadian Barcode of Life Network, and ROM Governor's Fund.

REFERENCES

- Banks J.C. & Paterson, A.M. 2007. A preliminary study of the genetic differences in New Zealand oystercatcher species. *N. Z. J. Zool.* **34**: 141-144
- Baker, A.J. 1973. Genetics of plumage variability in the variable oystercatcher (*Haematopus unicolor*). *Notornis*, **20**: 330-345
- Bray, T.C. & Hockey, P.A.R. 2015. Population genetics and demographic inferences in a recovering shorebird, the African Black Oystercatcher *Haematopus moquini*. *Ibis*, **157**: 171-176.
- Collar, N. J. & Stuart, S. N. 1985. *Threatened birds of Africa and related islands: the ICBP/IUCN Red Data Book*. International Council for Bird Preservation, and International Union for Conservation of Nature and Natural Resources, Cambridge, U.K.
- Gill, F. & Donsker, D. (eds). 2019. IOC World Bird List (v9.2). doi: 10.14344/IOC.ML.9.2.
- Hoang, D.T., Chernomor, O., von Haeseler, A., Minh, B.Q., & Vinh, L.S. 2017. UFBoot2: Improving the ultrafast bootstrap approximation. *Mol. Biol. Evol.* **35**: 518-52.
- Hayman, P., Marchant, J. & Prater, T. 1986. *Shorebirds: An identification guide to the waders of the World*. Helm: Bromley, UK.
- Helbig, A.J. & Seibold, I. 1999. Molecular phylogeny of Palearctic-African Acrocephalus and *Hippolais* warblers (Aves: Sylviidae). *Mol. Phylogenet. Evol.*, **11**: 246-260
- Herbert, P.D.N., Stoeckle, M.Y., Zemlak, T.S. & Francis, C.M. 2004. Identification of birds through DNA barcodes. *PLoS Biol.* **2**: 1657-1663
- Hockey, P.A.R. 1982. The taxonomic status of the Canary Islands Oystercatcher *Haematopus* (*niger*) *meadewaldoi*. *Bull. Brit. Orn. Club.* **102**: 77-83
- Hockey, P.A.R. 1987. The influence of coastal utilisation by man on the presumed extinction of the Canarian Black Oystercatcher *Haematopus meadewaldoi* (Bannerman). *Conserv. Biol.* **39**: 49-62.
- Inskipp, T.P., Collar, N.J., Mahood S.P. & Pilgrip, J.D. 2011. Species-level and other interesting changes suggested for Asian birds, 2010. *Birding ASIA*. **16**: 51-61
- Jehl, J.R. (1985) Hybridization and evolution of oystercatchers on the pacific coast of Baha California. *Ornithol. Mon.*, **36**: *Neotrp. Ornithol.*, 484-504
- Kalyaanamoorthy, S., Minh, B.Q., Wong, T.K.F., von Haeseler, A., & Jermiin, L.S. 2017. ModelFinder: Fast model selection for accurate phylogenetic estimates. *Nature Methods* 14:587–589
- Lavinia, P.D., Kerr, K.C.R., Tubaro, P.L., Hebert, P.D.N. & Lijtmaer, D.A. 2016. Calibrating the molecular clock beyond *cytochrome b*: assessing the evolutionary rate of COI in birds. *J. Avian Biol.* 47: 84–91
- Lee, M., Jeon, H.S., Choi, Y., Joo, S. & An, J. 2017. Complete mitochondrial genome of *Haematopus ostralegus* (Charandriiformes: Haematopodidae). *Mitochondrial DNA Part B* 2: 124-125
- Livezey, B.C. 2010. Phylogenetics of modern shorebirds (Charadriiformes) based on phenotypic evidence: analysis and discussion. *Zool. J. Linnean Soc.***160**: 567–618.
- Melville, D.S., Gerasimov, Y.N., Moores, N., Yat-Tung, Y & Bai, Q. 2014. Conservation assessment of Far Eastern Oystercatcher *Haematopus [ostralegus] osculans. International Wader Studies* 20: 129–154.
- Nguyen, L., Schmidt, H.A., von Haeseler, A., & Minh, B.Q. 2015. IQ-TREE: A fast and effective stochastic algorithm for estimating maximum likelihood phylogenies. *Mol. Biol. Evol.* **32**: 268-274
- Peters, J. L. 1934. *Check-list of the Birds of the World*. Volume 2. Harvard Univ. Press. Cambridge. Mass.

- Rando, J.C. & Alcover, J.A., 2010. On the extinction of the Dune Shearwater (*Puffinus holeae*) from the Canary Islands. *J. Ornithol.* **151**: 365-369.
- Shannon, T.J., McGowan, R.Y., Zonfrillo, B., Piertney S. & Collinson, J.M. 2014. A genetic screen of the island races of Wren Troglodytes troglodytes in the North-east Atlantic. *Bird Study* 61: 135-142

Stresemann, E. 1927. Die Schwartzen Austernfischer (Haematopus). Orn. Monatsh. 35: 71-77.

- Tavares, E. S., Senfeld, T., Shannon, T. J., Haddrath, O., Baker, A. J. & Collinson, J. M. Multi-loci coalescent analysis of Oystercatchers (*Haematopus*, Aves) suggests multiple events of morphological convergence. In prep.
- Theron, E. Hawkins, K., Bermingham, E., Ricklefs, R.E. & Mundy, N.I., The molecular basis of an avian plumage polymorphism in the wild: A melanocortin-1-receptor point mutation is perfectly associated with the melanic plumage morph of the bananaquit, *Coereba flaveola*. *Curr. Biol.* 11: 550-557.
- Trifinopoulos J, Nguyen LT, von Haeseler A, Minh BQ 2016. W-IQ-TREE: a fast online phylogenetic tool for maximum likelihood analysis. *Nucl. Acids Res.* 44 (W1): W232-W235.
- Uy, J.A.C.& Vargas-Castro L.E. 2015. Island size predicts the frequency of melanic birds in the color-polymorphic flycatcher *Monarcha castaneiventris* of the Solomon Islands. *Auk* **132**: 787-794.
- Vaurie, C. 1965. *The Birds of the Palearctic Fauna: A Systematic Reference. Non-passeriformes.* Witherby: London.
- Valledor de Lozoya, A. 2013. *Ostrero Canario: Historia y Biología de la Primera Especie de la Fauna Española Extinguida por el Hombre*. Editorial Ministerio de Agricultura, Alimentación y Medio Ambiente.
- Van Grouw, H. 2017. The dark side of brids: melanism facts and fiction. Bull. B. O. C. 137:12-36
- Weir, J.T. & Schluter, D. 2008. Calibrating the avian molecular clock. Mol. Ecol. 17: 2321-2328.
- Wolters, H. E. 1975-1982. Die Vogelarten der Erde. Eine systematische List emit Verbreitungsangaben sowie deutschen und englischen Namen (7 volumes). Paul Parey, Hamburg & Berlin.

Table 1. Details of samples used in this study and Genbank accession numbers.

Sample code	Species	Tissue type	Origin	СОІ	cyt b	NADH2
HO03	H. ostralegus	feather	Scotland, UK	LR595929	LR595930	LR595931
HMo01	H. moquini	blood	Cape Town, South Africa	LR595932	LR595933	LR595934
HMo09	H. moquini	blood	Bettys Bay, South Africa	LR595935	LR595936	LR595937
HMo17	H. moquini	blood	Jutten Island, South Africa	LR595938	LR595939	LR595940
HMo19	H. moquini	blood	Jutten Island, South Africa	LR595941	LR595942	LR595943
HMo30	H. moquini	blood	Cape Town, South Africa	LR595944	LR595945	LR595946
HMo32	H. moquini	blood	Cape Town, South Africa	LR595947	LR595948	LR595949
HMeL01 (Liverpool T.16000)	H. meadewaldoi	toepad	La Graciosa, Canary Islands (Spain)	LR595950	LR595951	Figure S5
HMeM01 (Manchester B.9162)	H. meadewaldoi	toepad	Jandia, Fuerteventura, Canary Islands (Spain)	LR595952	LR595953	Figure S5
Mead5 (NHMUK.19 05.12.22.322)	H. meadewaldoi	toepad	La Graciosa, Canary Islands (Spain)	LR595954	-	-
Mead6 (NHMUK.19 39.12.9.25)	H. meadewaldoi	toepad	La Graciosa, Canary Islands (Spain)	LR595955	-	-
Hx01 NHMUK 1938.11.15.1	unknown	toepad	The Gambia	Figure S5	Figure S5	Figure S5

Figures

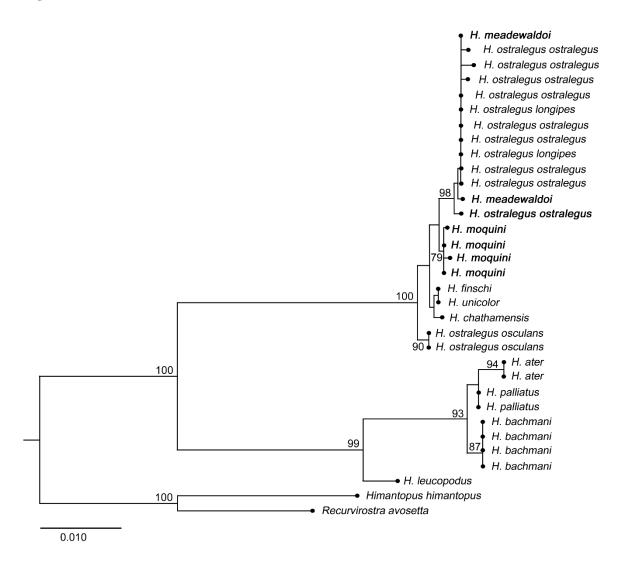


Figure 1. Phylogeny of oystercatchers *Haemotopus*. Maximum likelihood tree based on alignment of concatenated *COI* and *cytb* sequences of individuals sequenced in this study (bold), with additional sequences of all taxa downloaded from Genbank. Bootstrap support for key nodes are shown. A cladogram based on the same alignment, with Accession numbers other identifiers is presented in Supplementary Material. The *COI* sequences obtained from NHM specimens were identical to those of the Liverpool and Manchester specimens not included in this figure.

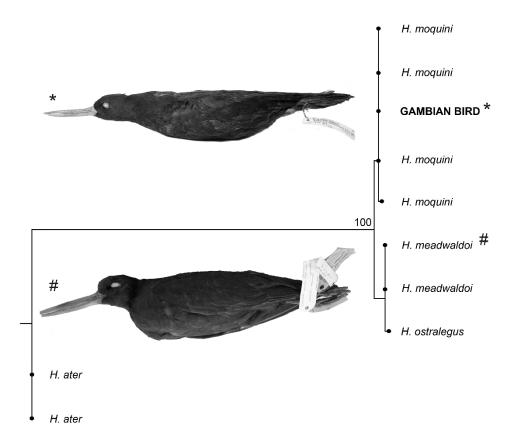


Figure 2. Identification of the mystery Gambian oystercatcher as an African Oystercatcher. Maximum likelihood tree based on alignment of 281 bp of concatenated fragments of *COI*, *cytb* and *NADH2* sequences of African Canary Islands and Eurasian Oystercatchers sequenced in this study (bold), with additional sequences of outgroup Blackish Oystercatcher downloaded from Genbank. A cladogram based on the same alignment, with full bootstrap values, Accession numbers other identifiers is presented in Supplementary Material. Thumbnail images of the Gambian bird (top) and Canary Islands Oystercatcher from Liverpool World Museum (bottom) are presented. See also Supplementary Figures 1, 2, 4, 5. Image credits: top Lucie Goodayle/NHM; bottom Alexander C. Lees.