

## **INTEGRATION OF MITOCHONDRIAL AND GENOMIC DATA FOR** THE INFERENCE OF CURRUCA MELANOCEPHALA PHYLOGEOGRAPHY



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The Western Palearctic (WP) has always been a focus of phylogeographic studies, especially given the peculiarities of its biogeographic regions. In addition, the Plio-Pleistocene ice ages and the glacial refugia have played an undeniable role in influencing the species fluctuations and genetic arrangements. Although the avian phylogeography has been extensively studied in the WP [1], few studies have focused on species originated and persisted in the Mediterranean basin.

The Sardinian Warbler (Curruca melanocephala) is a largely sedentary passerine belonging to the Curruca clade (ex Sylvia), a genus within the Sylviidae family. Currently, 4 subspecies are recognised, based on distribution and morphological characteristics [2,3, Fig. 1]: i) C. m. melanocephala; ii) C. m. leucogastra (Canary Islands); iii) C. m. momus (Levant); iv) C. m. valverdei (Western Sahara).



This study aims to investigates the evolutionary path of the species, whether or not it confirm the subpspecies classification and the reasons behind the current genetic arrangment.

[1] Pârâu LG, Wink M, (2021). J Ornithol.;162(4):937-959 [2] del Hoyo J, Collar N.J. (2016). Lynx Edictions [3] Cabot J, Urdiales C, (2005). B.O.C.;125(3):165-175

A total of 96 blood/tissue samples were analysed, covering most of the species' range (Fig. 2). The samples came from the UPO Biobank collection, several museum repositories and ringing stations. **DNA** was extracted with specific kit and a **polymerase** chain reaction (PCR) was performed to amplify several mtDNA markers (i.e. cox1, cytb, nd2). Sanger sequencing was then performed to obtain the sequences of the markers. In addition, single-digest RAD libraries were prepared and sequenced to characterise genome-wide SNPs. Bioinformatic and phylogenetic analyses were performed on both datasets using different software (i.e. Samtools, STACKS, RStudio, MrBayes). In addition, species distribution models were predicted using the R package "smd", based on occurrence data obtained from GBIF and bioclimatic variables obtained from the WorldClim database.

Fig. 3: (A) phylogenetic tree reconstruction of the mtDNA concatenated dataset and MJ network of the cytb dataset; (B) ADMIXTURE analysis on the genome SNPs; (C) current and at Last Glacial Maximum SDM of Curruca melanocephala

CONCLUSIONS



Fig. 2: Sampling sites

- A weak geographic structure was indicated by the presence of widely distributed haplogroup across the species range. An exception was found for Israeli samples, which, in addition to the morphology, validates the C. m. momus subspecies. An entangled arrangement with blurred geographical lineages was delineated for *leucogastra* and *valverdei*;
- The SNPs analysis confirms the results of mtDNA: a case of genetic admixture is evident among geographically disjunct populations, except for the Israeli (*C. m. momus*) individuals, which share less ancestry with the other populations;
- The species distribution model under at the Last Glacial Maximum bioclimatic conditions emphasised that populations were distributed along a reduced but continuous range and were therefore able to interbreed.

Overall, our results suggest that an uninterrupted range during the glacial period allowed continuous gene flow between populations, leading to the current panmixia. Further sampling efforts in additional areas, including biometric analysis and bioacoustics, may shed additional light on the phylogenetic history of this species.

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