



News & Views

Y-chromosome Peruvian origin of the 500-year-old Inca child mummy sacrificed in Cerro Aconcagua (Argentina)

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A frozen mummy was found in Cerro Aconcagua (Argentina) in 1985 (Fig. 1a–c). Archaeologists identified this mummy as a seven-year-old Inca sacrifice victim living at the time of the Inca Civilization about 500 years ago [1]. The child was sacrificed following an Incan ritual known as *capacocha*. The complete mitochondrial DNA (mtDNA) genome of this mummy was recently analyzed in Gómez-Carballa et al. [2]. This mitogenome belongs to a new sub-branch of the Native American phylogeny named C1bi dated to approximately 14 thousand years ago (kya). Information retrieved from a large database of mtDNA profiles indicated the existence of a few closely related haplotypes in Peru (including one haplotype retrieved from ancient DNA of an individual belonging to the Wari Empire) and the Aymaras from Bolivia. Overall these data suggested a Peruvian Inca origin for this C1bi haplotype, in good agreement with archaeological reports [1].

There is extensive literature on Y-chromosome studies in America, e.g. [3–8]. To the best of our knowledge, only a few Y-chromosome studies have been carried out to date on ancient or historical DNA from human remains of Pre-Columbian South American specimens, e.g. [9,10]; none of them targeted Native American mummies. The present study aimed at analyzing the Y-chromosome profile of the mummy of the Cerro Aconcagua, infer its most likely geographic origin, and understand the findings in the light of previous anthropological, archaeological and genetic research.

A total of 14 Y-SNPs were successfully genotyped in the mummy's DNA (see online supplementary text for technical information). The Y-SNP haplotype could be assigned to haplogroup Q-M3. All markers except M3 were ancestral (Supplementary text and Fig. S1 online). The SNP status of the mummy Y-chromosome is also consistent with the SNP information provided by the Y-chromosome Reference Database (YHRD; <https://yhrd.org>), indicating that its Peruvian 1-step mutation neighbor haplotype was allocated to Q1a2-M3.

The full PP23 Y-STR panel was successfully genotyped in the mummy's DNA (Supplementary text) with the following allele combination: 15–14–25–31–16–14–16/16–13–10–12–22–14–12–14–11–20–16–24–13–11–16–10 (the alleles referring to the following Y-STRs: DYS456, DYS389I, DYS390, DYS389II, DYS458, DYS19, DYS385a/b, DYS393, DYS391, DYS439, DYS635, DYS392, YGATAH4, DYS437, DYS438, DYS448, DYS576, DYS481, DYS549, DYS533, DYS570, DYS643; Fig. S2 (online). The mummy was additional genotyped for the Yfiler Y-STR panel, and the genotypes for the common Y-STRs in the two kits matched (Supplementary text and Fig. S3 online). Initial database searches were carried in the YHRD (Release R55), a database containing 39,414 PPY23 profiles worldwide recorded so far, including Argentinean, Peruvian and other South American populations [11]. The mummy's haplotype was unique in the YHRD (query sent in March 2018). In order to facilitate the search for closer phylogenetically related profiles of the mummy's haplotype in present-day populations, we collapsed the PP23 haplotype into the minimal haplotype (MHT: DYS19–DYS389I–DYS389II–DYS390–DYS391–DYS392–DYS393–DYS385) and performed a new search in the YHRD (query sent in March 2018). Again, no exact matches were found ($n = 197,102$ worldwide haplotypes; 2,996 classified as Native American). We finally searched for 1-step mutational neighbors in this database of minimal haplotypes. Three 1-step haplotypes emerged from this search: one in Peru ($n = 703$), and two in admixed population from United States ($n = 8,848$). Fig. S4 (online) shows the Heat Map of the 1-step mutation mummy haplotypes.

We carried out additional searches in a large database of Native American haplotypes compiled from the literature (hereafter NADB, coverage = 0.44; Supplementary text; Table S1 and Fig. S5 online). We did not find any additional matches for the mummy's haplotype. Furthermore, we did not find closely related haplotypes (1-step mutational profiles) to the mummy's one. We observed however that on average, the haplotypes contained in the NADB have 1.76 matches within this database, and 1.84 1-step mutational neighbors. Therefore, the fact that the mummy's haplotypes has nor matches nor mutational neighbors

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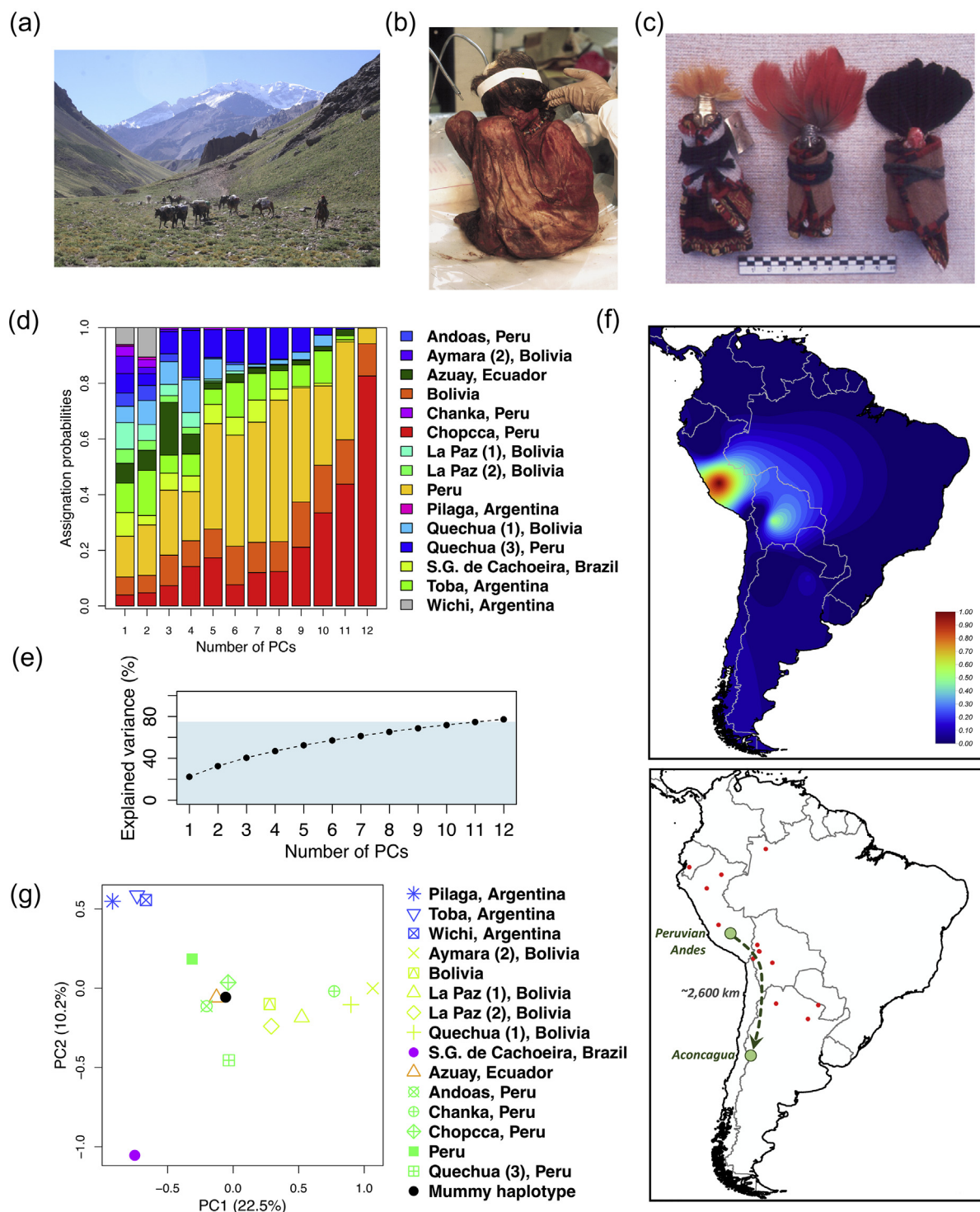


Fig. 1. Photos of the mummy and statistical/population analyses carried out on the mummy's Y-STR haplotype. (a) Photo of the Cerro Pirámide in the Aconcagua mountain: in 1985 a sacrificed seven-year-old boy wrapped in textiles and surrounded by statuettes was found at the base of the Cerro Pirámide. (b) Photo taken of the unwrapped mummy when carrying out the preliminary studies. (c) Photo of three out of the six statuettes found in the mummy's burial. (d) Probabilistic classification of the mummy's Y-chromosome haplotype in South American Native American samples as a function of the number of principal components (PCs) added to the algorithm. (e) Evolution of the accumulated informativeness as more PCs are added, until reaching 75% of the variation explained. (f) Map (top) representing the interpolated probabilities of classification (the values correspond to the data represented in (c) for the first 12 PCs), and map (bottom) indicating the sampling points plus the Aconcagua site and the most likely geographic origin of the Y-STR profile. (g) PCA computed on the mummy's haplotype and the set of South American populations considered in the NADB. The labels of the South American sample sets in (d) and (g) correspond to those in Table S1. The photos in (a) to (c) were taken and provided by one of the co-authors of the present study (J.R.B.) who was involved in the preliminary anthropological studies carried out on the Aconcagua mummy.

indirectly suggests that it represents a particularly rare lineage within the pool of Y-chromosomes of contemporary South American populations. In addition, no exact matches could be found in an additional search carried out on the US Y-STR database release

4.2 ($n = 35,660$; query sent in January 2018; <https://www.usystrdatabase.org>).

Estimation of the most likely geographical origin of the mummy's haplotype was initially carried out on a continental scale

using a large continental database (hereafter CDB; Supplementary text; Table S2 and Fig. S6 online). The haplotype of the mummy clearly classifies into the Native American metapopulation with a probability > 0.999 and using the information accounted for by the first 29 PCs (Table S3 online). The classification into the Native American group is already robust ($P = 0.772$) using only the first 10 PCs (data not shown).

Next, we classified the mummy's haplotype into the NADB. The population sample yielding the highest probability was the Choppca from Huancavelica ($P = 0.827$) [12], a Native Quechua-speaking ethnic group inhabiting the highlands of Peru (Fig. 1d). The amount of total variation explained by the first 11 PCs is >75% of the cumulative PCA variation (Fig. 1e). Fig. 1f shows the interpolated probabilities (for the 75% of the variation accounted for in the classification algorithm) of the mummy's Y-profile into the 15 Native American populations used for classification. The map highlights the most likely origin of the mummy's haplotype in the heart of the Peruvian Andes.

The classification algorithm was run again on the NADB, excluding the Choppca from the database. This time, the population sample that scored the highest probability was another sample from Peru ($P = 0.692$), (Fig. S7 online).

A final classification run was carried out between the two main linguistic groups from the Andes, the Quechua and the Aymara. The algorithm classifies the mummy's haplotype into the Quechua cluster with a high probability ($P > 0.999$).

A PCA was computed to further explore the affinities of the mummy's haplotype to populations in South America (those considered in the NADB); (Fig. 1g). PC1, which accounts for 22.5% of the variation, is very informative in revealing the close proximity of the mummy's haplotype to a main Peruvian cluster. The neighboring Andean region of Azuay in Ecuador [13], where the predominant Native American language is the Kichwa (Quechua) also falls in this group of samples. PC2, which accounts also for a substantial amount of variation (10.2%) reveals again the proximity of the mummy's haplotype to this Peruvian cluster and to the Bolivian sample sets (Fig. 1g).

Remarkably, the mummy was found in the Argentinean Andes, about 2,600 km southwards to the most likely origin of its haplotype. Overall, the data are consistent with the hypothesis that the child could have been removed from his birth region (Peru) and taken far away to the sacred place of the Aconagua (Argentina) for sacrifice. This hypothesis fits also with archaeological and anthropological findings [1]. Supportive evidence favoring a Peruvian origin for the mummy is the fact that the mummy haplotype has more affinities to the Y-chromosome profiles that represent the Quechua linguistic group than to the Aymara. It is known that the Quechua probably originated around the Cuzco region, and it was the official language of the Incas [14] at the time of the Aconagua mummy and the Spanish conquest. The Inca Civilization is in fact known as the Quechua civilization. Peru is presently the region with the main Quechua speaking community of the Andes; while the Aymara is the main language in neighboring Bolivia. A Peruvian origin of the mummy's haplotype would also add to the genetic evidence indicating the existence of demographic movements along the Pacific Coastline during the Inca Period, a migration route that was already important since the times of the initial South American settlement by the first Paleoindians [15–17].

Summarizing, the phylogeographic Y-chromosome data and the statistical analyses carried out are consistent with the mtDNA findings [2], and the anthropological and archaeological evidence, which altogether agree well with a Peruvian Inca origin for the Aconagua mummy. The affinities between the Y-chromosome profile to the profiles characterizing the Quechuan cluster add further support to this hypothesis. Overall, the genetic evidence is also consistent with the archaeological hypothesis claiming that the Aconagua boy could have been carried > 2,600 km South from

its origins in the Peruvian Andes for sacrifice in the sacred Aconagua mountain.

Acknowledgment

We are indebted to Lutz Roewer and Maria Geppert for providing information on the Y-SNPs of the mummy.

Conflict of interest

The authors declare that they have no conflict of interest.

Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at <https://doi.org/10.1016/j.scib.2018.08.009>.

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