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Ancient Human Mitogenome of the Beagle Channel (Tierra del Fuego): An Argentine Collaborative Project

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Abstract

The increasing use of massively parallel sequencing in the study of current and ancient human populations has enabled new approaches to bioanthropological and archaeological issues; however, its application to archaeological samples requires the use of technologies that are not easily accessible outside US and European research centers. To obtain an ancient mitogenome in Argentina, several institutions collaborated to apply massively parallel sequencing and bioinformatic methodologies on an enriched ancient DNA library of an individual from the Beagle Channel (dated 1504 ± 46 years BP), a region of particular interest for this line of inquiry. Phylogenetic reconstruction showed a close relationship with a Yamana from Navarino Island and an individual from Hoste Island (Chilean Antarctic Province): the three shared an ancestor who lived between 203 and 4,439 years ago. These three have mutations reported only for current and ancient individuals from the Beagle Channel, and their relationship with the rest of the D1g sub-haplogroups is unclear. The results obtained here are consistent with the reduction of mobility in the Fuegian archipelago around 4500 years BP that has been proposed based on archaeological evidence.

Resumen

La creciente utilización de la secuenciación masiva en el estudio de poblaciones humanas actuales y antiguas, permitió nuevos abordajes a problemáticas bioantropológicas y arqueológicas. Sin embargo, su aplicación en muestras arqueológicas requiere del uso de tecnologías que no son fácilmente accesibles fuera de los centros de investigación estadounidenses y europeos. Con el fin de obtener un mitogenoma antiguo realizado en Argentina, varias instituciones colaboraron para aplicar metodologías de secuenciación masiva y bioinformática sobre una biblioteca enriquecida de ADN, de un individuo del canal Beagle (datado en 1504 ± 46 años AP), región cuyas singularidades la vuelven un lugar de particular interés para esta línea de estudio. Las reconstrucciones filogenéticas realizadas lo muestran cercanamente emparentado a un yamana de la Isla Navarino y a un individuo de la isla Hoste (Provincia de la Antártica Chilena), compartiendo los tres un ancestro de 203–4,439 años atrás. Éstos poseen mutaciones reportadas únicamente para

individuos actuales y antiguos del canal Beagle, y su relación con el resto de los subhaplogrupos de D1g es incierta. Los resultados obtenidos son consistentes con la reducción de la movilidad en el archipiélago fueguino propuesta en base a evidencias arqueológicas hacia 4500 años AP.

Keywords: ancient DNA; mitogenome; Tierra del Fuego; D1g

Palabras clave: ADN antiguo; mitogenoma; Tierra del Fuego; D1g

Over the last few decades, bioanthropology and archaeological research has incorporated the use of genetic information recovered from skeletal remains to examine a variety of issues regarding the origin and worldwide dispersion of our species. DNA studies on past and current populations have been at the center of the latest models of occupation of the Americas and of the different microevolutionary processes shaping the genetic diversity of native populations (e.g., Brandini et al. 2017; Lindo et al. 2018; Perez et al. 2016; Posth et al. 2018). The use of massively parallel sequencing has facilitated the recovery of mitochondrial and nuclear genomes, enabling increasing precision in the definition of American founding mitochondrial lineages, the description of regional peopling processes and interpopulation dynamics, the identification of adaptations to local environments, and changes in demographic patterns (De la Fuente et al. 2018; Fehren-Schmitz and Georges 2016; Lindo et al. 2018; Posth et al. 2018).

However, the application of such procedures to archaeological samples poses challenges and requires strategies that cannot be readily implemented by most researchers because of their high cost or the lack of adequate infrastructure, equipment, and specialized technical staff; one such difficult-to-implement strategy is the fabrication of special probes to capture the target DNA and the enrichment of sequencing libraries. Therefore, analyses on South American archaeological samples are usually conducted in the context of cooperation agreements between local researchers and nonlocal research centers mostly located in the United States and Europe (Wade 2018). As a result, the involvement of South American researchers in ancient DNA (aDNA) studies is frequently subordinated to the research agenda of the nonlocal laboratories. This situation discourages the pursuit of locally relevant research objectives that often involve more restricted time or spatial scales (Arencibia et al. 2019; Cardozo et al. 2021; Crespo et al. 2020; Motti et al. 2020; Postillone et al. 2020; Russo et al. 2016, 2018; Tavella et al. 2020). The development of such asymmetrical relationships is common to many scientific fields that depend on costly technological development; this “subordinate integration” (Kreimer 2006; Kreimer and Ugartemendía 2007) is a recurring phenomenon in the global division of scientific labor today. In this context, the procurement of resources to support the pursuit of avenues of research that are consistent with local and regional goals is essential to the local advancement of archaeological science.

The Beagle Channel area holds a particular interest for genetic studies. The area was probably one of the last territories to be reached by humans, which is one reason why it has been intensively studied over many years by different research groups. These studies have yielded a large and diverse body of information that has provided valuable contextual information for framing genetic studies. The earliest archaeological signal in the region dates to about 8000 years BP (Zangrando et al. 2018, 2022) and the available zooarchaeological, technological, and isotopic evidence points to the existence of maritime adaptations in the area as early as 6400 years BP (Orquera and Piana 1999, 2009; Suby et al. 2011; Zangrando 2009). Additionally, Patagonia in general has exceptional preservation conditions that facilitate the recovery of genetic material, a fact made evident by the increasing number of Patagonian aDNA analyses performed in laboratories around the world (Arencibia et al. 2019; Crespo et al. 2017a, 2017b, 2020; de la Fuente et al. 2015, 2018; Lalueza et al. 1997; Motti et al. 2015, 2019; Nakatsuka et al. 2020; Parolin et al. 2019; Raghavan et al. 2015).

From a genetic point of view, maternal lineage C1 has been found in high frequencies in the Beagle Channel area (Tierra del Fuego, Argentina; Figure 1) and D1g and D4h3a have also been identified, albeit in lower proportions (Crespo et al. 2017b, 2018; de la Fuente et al. 2015, 2018; Lalueza Fox 1996). Similar frequencies of the C1 and D1g lineages have also been observed in ancient samples

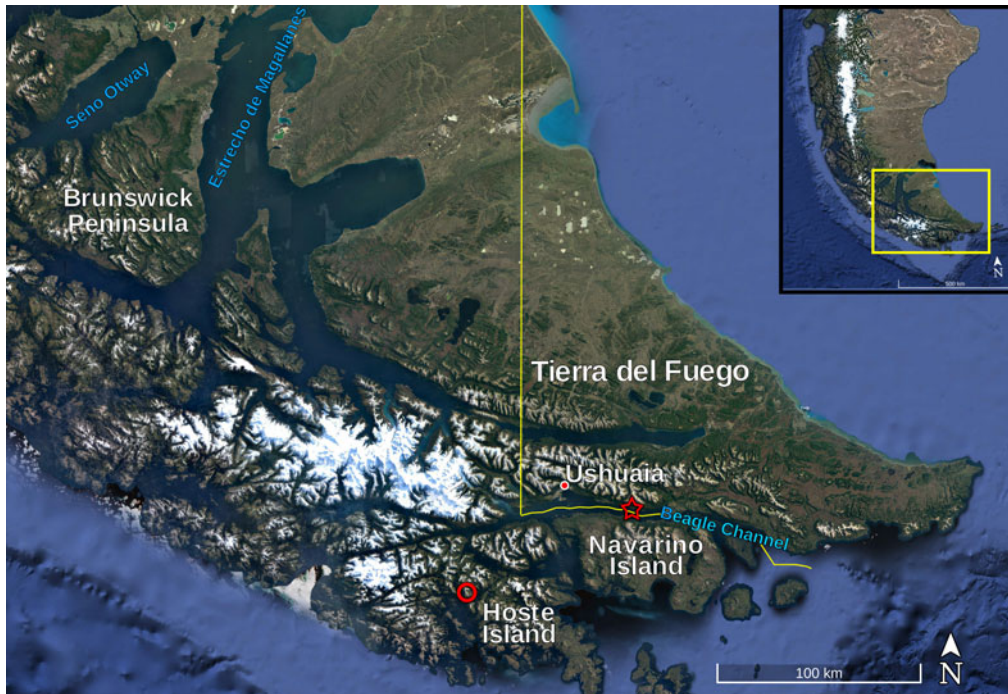


Figure 1. Region of interest. The star indicates the location of the Paiashauaia 1 burial by the Beagle Channel (Tierra del Fuego, Argentina); the circle indicates the location of the IPY08 burial by Seno Año Nuevo (Chile; de la Fuente et al. 2018). Image generated with Google Earth (v7.3.3.7786).

from nearby areas, such as Peninsula Mitre and Isla de los Estados; these findings could indicate different degrees of contact or shared origins among diverse native groups in Tierra del Fuego and the Beagle Channel (Crespo et al. 2017b, 2020; Nakatsuka et al. 2020).

Given the region's distinctive characteristics, we studied an individual recovered from an archaeological site located on the Beagle Channel coast by analyzing its lineage within the regional phylogenetic context. To this end, we implemented a process to build and enrich an aDNA library and to undertake the massively parallel sequencing methodologies and bioinformatic procedures that are necessary to obtain a mitogenome “made in Argentina” (i.e., only using locally available resources).

Materials and Methods

The Analyzed Individual

An upper left incisor was taken from human remains recovered from Paiashauaia I, a primary mortuary context located at a rocky outcrop next to the mouth of the Paraná River, a few meters from the Beagle Channel coast (Figure 1; laboratory code: PZ3). Bioarchaeological analysis determined that the individual was a female between 35 and 45 years old, dated at 1504 ± 46 years BP by radiocarbon AMS (Suby et al. 2011; for more details on methodology, see Supplemental Text 1). These remains are currently preserved in the Museo del Fin del Mundo's repository in Tierra del Fuego; permits for sample transportation and analysis were obtained from the Secretaría de Cultura of Tierra del Fuego in 2016. In addition, the sample was treated with respect and scientific rigor as set forth by the code of ethics for the study of human remains of the Asociación de Antropología Biológica of Argentina (2007; Aranda et al. 2014).

The hypervariable regions 1 and 2 of PZ3's mitochondrial DNA were successfully recovered, amplified, and sequenced by Sanger (Crespo et al. 2020). According to this study, PZ3's mitochondrial lineage was a distinctive D1g exclusively found in the Beagle Channel area.

Laboratory Protocols

All precautions for preventing and detecting exogenous contamination in the ancient sample were followed (see the detailed protocol in Supplemental Text 1). DNA extraction and library preparation were carried out at the aDNA laboratory clean room facility at the Universidad Maimónides in Buenos Aires. Library amplification, bait preparation, and capture were done at the laboratories of the Instituto Nacional de Tecnología Agropecuaria (INTA). The sample was powdered for DNA extraction. Extraction blank controls were conducted and subjected to PCR for the hypervariable region, along with the archaeological samples, to monitor for possible contamination.

The aDNA library was constructed using the NEBNext Ultra II DNA Library Prep Kit for Illumina following the manufacturer's instructions (Supplemental Text 1). DNA fragmentation was not performed before construction of the library. After USER enzyme (NEB) treatment, the adaptor-ligated DNA was purified without size selection using AMPureXP Beads (Beckman Coulter).

The indexed aDNA library was captured in solution using 3' biotin-labeled probes with the mitochondrial genome of an H haplogroup blood donor, coupled to Dynabeads® M-280 Streptavidin Magnetic Beads (Invitrogen; Maricic et al. 2010); the solution was incubated at 55°C for 48 hours. After washing, the captured library was eluted by heating, and the DNA was purified (Supplemental Text 1).

The captured aDNA library was sequenced by synthesis with MiSeq Reagent Kit v2 (2 × 150) at INTA's Consorcio Argentino de Tecnología Genómica (CATG). PCR and Sanger sequencing were used to confirm SNPs in two low-depth regions (primers specified in Supplemental Table 1).

Bioinformatic Analysis

Processing and Mapping of Sequenced Reads and Variant Calling. The quality of the Illumina reads was evaluated with FastQC (v0.10.1), and the reads were trimmed and end-clipped to a PHRED score of 30 using Cutadapt (v1.16; Martin 2011; see the detailed protocol in the Supplemental Text 1). Paired-end reads were then merged using AdapterRemoval (v2.3.3; Schubert et al. 2016) and mapped to the revised Cambridge Reference Sequence (rCRS; Andrews et al. 1999) using the Burrows Wheeler Aligner (BWA; v0.7.10; Li and Durbin 2009). The output was converted to a BAM file using Samtools (v1.7; Li and Durbin 2009), and one 3' nucleotide was trimmed in the BAM file with bamUtil (v1.0.15; Jun et al. 2015). Mapping statistics were calculated with Bamtools (v2.5.1; Barnett et al. 2011) and Samtools, duplications were marked and discarded using Picard-tools (v2.18; <http://broadinstitute.github.io/picard/>), and aDNA damage patterns were assessed using MapDamage2 (v2.0; Jónsson et al. 2013).

Variant calling was performed using BCFtools (v1.9; Danecek and McCarthy 2017; Narasimhan et al. 2016). The information yielded by the Sanger sequences and obtained for HVR1 in Crespo and colleagues (2020) was incorporated into variant calling. Finally, variant filtering was also made using BCFtools.

Construction of the Consensus Mitogenome. A final VCF file was used to construct a consensus genome of the individual PZ3 using BCFtools (Supplemental Text 1). Positions with depth <2 were treated as missing data (N). The mitochondrial haplogroup was determined by using HaploGrep2 (v2.2; Weissensteiner et al. 2016).

Haplotype Polymorphisms and Phylogenetic Relationships

We compiled a database that included ancient and modern D1 sequences from Argentina, Brazil, and Chile to compare the PZ3 mitochondrial haplotype to other D1 mitogenomes. Sequences reported up to 2016 were obtained from Phylotree Build 17 (February 18, 2016; van Oven and Kayser 2009), and more recently reported sequences were retrieved from the literature. One representative of each of the D1 sublineages not reported in the three countries was also included for analysis, as well as one D4h3a and the RSR5 (Behar et al. 2012), yielding a final data matrix of 79 sequences (Supplemental Table 2).

Sequences were aligned using the FFT-NS-2 algorithm in MAFFT (v7.471; Katoh et al. 2002). The absolute number of changes among haplotypes was computed in R (v3.4.4; R Development Core Team 2020) using the haplotypes package (v1.1.2; Aktas 2020) while considering gaps as a fifth character

state. A haplotype median-joining network was constructed using Network (v10.2; <http://www.fluxus-engineering.com>). To facilitate visualization, only the D1 individuals from Argentina and Chile were included in the analysis. Each polymorphic site was weighted according to the mutational rates described by Soares and colleagues (2009). We did not consider mutations 309.1C, 315.1C, A16182c, A16183c, and 16193.1C, or positions with gaps and missing values.

The phylogenetic analysis of D1 mitogenomes was performed with BEAST (v2.6.2; Bouckaert et al. 2019). Two partitions were considered, one corresponding to the HVR (1–577; 16023–16569) and the other to the coding region (578–16022). Site models were averaged using bModelTest (Bouckaert and Drummond 2017). As suggested for intraspecific data (Drummond and Bouckaert 2015), analyses were conducted under a strict molecular clock model, with the prior distribution for the clock rate set as the default. Analyses were done under the coalescent constant population, coalescent exponential population, and coalescent extended Bayesian skyline models. The MCMC chain length was set to 15 million, 20 million, and 65 million iterations, respectively, and data were stored every 1,500th iteration. A clock rate of 0.024694 substitutions per site per million years (Llamas et al. 2016) and a ploidy of 0.5 were set for the coalescent extended Bayesian skyline. The subsequent model selection was conducted using nested sampling (Maturana Russel et al. 2019). For the Bayesian skyline plot, the y-axis transformation to effective population size was performed using a generational time of 25 years (as used in Brandini et al. 2017).

The run parameters were checked with Tracer (v1.7.1; Rambaut et al. 2018), and trees maximizing the posterior probability were scored with TreeAnnotator (v2.6.2; Bouckaert et al. 2019). A qualitative analysis of the consensus trees obtained was performed with DensiTree (v2.2.7; Bouckaert et al. 2019). Graphical postprocessing was done with FigTree (v1.4.4; Rambaut 2018), and Inkscape (v1.0; www.inkscape.org) was used for image design.

Results

No sign of exogenous DNA contamination was found in the extraction blank, and we ruled out any contamination of the sample with DNA from the researchers who had contact with it. The results of concentration, quantification, and fragment analyses indicate that the library was successfully generated. There are typical postmortem DNA damage patterns that help distinguish ancient sequences from modern contaminants, such as short sequences and cytosine deamination, observed as transitions from C to T at the 5' end and from G to A at the 3' end of the readings (Briggs et al. 2007; Jónsson et al. 2013; Sawyer et al. 2012). Supplemental Figure 1 shows the results obtained with MapDamage2. The excess of purines at the position immediately before the fragment start is a characteristic pattern in ancient DNA (Supplemental Figure 1A); Supplemental Figure 1B illustrates what is expected for a library treated with the USER enzyme, which removes most of these signs of damage.

About 43% of the reads from the enriched sequence PZ3 library passed bioinformatic filters and were mapped against the rCRS (Supplemental Table 3). The average depth achieved was 8X. The incorporation of Sanger sequences allowed us to increase the read depth for two variant positions. The generated consensus mitogenome had 99.7% coverage. A total of 37 SNPs were identified in the haplotype of individual PZ3 with respect to the rCRS (Table 1). Both this number of mutations and their distribution pattern (coding region: 27 mutations / 15,449 total bases; HVR: 10 mutations / 1,120 total bases) were consistent with expected values (Soares et al. 2009), giving robustness to the results.

This haplotype was assigned to the haplogroup D1g and is very similar to two previously reported individuals from the Chilean Antarctic Province: YA2D (de Saint Pierre et al. 2012) and IPY08 (De la Fuente et al. 2018). These three haplotypes are characterized by the lack of the A8116 G mutation, originally proposed as a defining characteristic of the D1g haplogroup along with the C16187T reversion (Bodner et al. 2012) but that was later removed in the clade redefinition of de Saint Pierre and colleagues (2012). The only differences between these and PZ3 are located at positions 309–315 and 3107 (Figure 2 and Supplemental Table 4), which are either hotspot regions or challenging positions for aDNA reconstruction and thus have low or no evolutionary weight.

Table 1. Variants Found in the PZ3 Consensus Mitogenome.

Position	Substitution (rCRS>PZ3)	Location	Aminoacid change
73	A>G	HVR-2	—
263	A>G	HVR-2	—
489	T>C	HVR-3	—
750	A>G	12S	—
1438	A>G	12S	—
2092	C>T	16S	—
2706	A>G	16S	—
3010	G>A	16S	—
4769	A>G	ND2	Ile-Met
4883	C>T	ND2	Pro-Pro
5178	C>A	ND2	Leu-Ile
7028	C>T	COX1	Ala-Ala
8414	C>T	ATP8	Leu-Phe
8701	A>G	ATP6	Thr-Ala
8860	A>G	ATP6	Thr-Ala
9540	T>C	CO3	Leu-Leu
10202	C>T	ND3	Arg-Arg
10398	A>G	ND3	Thr-Ala
10400	C>T	ND3	Thr-Thr
10724	T>C	ND4L	Tyr-Tyr
10873	T>C	ND4	Pro-Pro
11719	G>A	ND4	Gly-Gly
12705	C>T	ND5	Ile-Ile
13020	T>C	ND5	Gly-Gly
14668	C>T	ND6	Met-Ile
14766	C>T	CYTB	Thr-Ile
14783	T>C	CYTB	Leu-Leu
15043	G>A	CYTB	Gly-Gly
15301	G>A	CYTB	Leu-Leu
15326	A>G	CYTB	Thr-Ala
16086	T>C	HVR-1	—
16187	C>T	HVR-1	—
16189	T>C	HVR-1	—
16223	C>T	HVR-1	—
16286	C>T	HVR-1	—
16325	T>C	HVR-1	—
16362	T>C	HVR-1	—

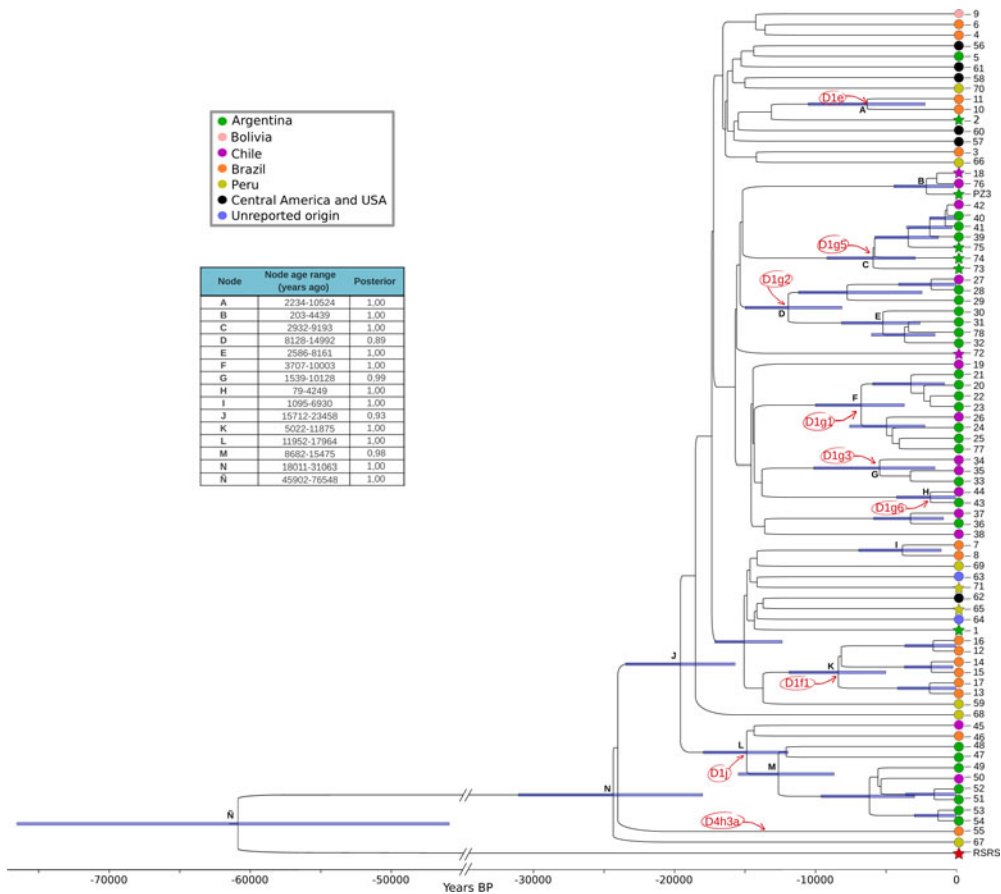


Figure 2. Bayesian phylogeny depicting the relationships between individuals of the human mitochondrial haplogroup D1 and external groups. The x-axis denotes time into the past in years. The tree is based on an extended Bayesian skyline analysis of the mitogenomes considering two partitions (coding and hypervariable region) performed in BEAST (v.2.6.2; Bouckaert et al. 2019). Sequence sources and sample numbers are available in Supplemental Table 2. Nodes with posterior probability >0.7 have bars indicating age ranges accumulating 95% posterior probability. The table shows the age ranges and posterior probability for each node. Names of major well-supported sub-haplogroups are indicated. Circles: modern individuals; stars: ancient individuals.

The geographic information of the individuals included in the database (Supplemental Table 2) made possible the Bayesian phylogenetic reconstruction of the mitochondrial D1 lineage for Argentina, Brazil, and Chile. Nested sampling results pointed to the extended Bayesian skyline as the best of the three models considered (ML: -25,739.289; SD: 16.571; Figure 2). The topology obtained strongly suggests that PZ3 is closely related to YA2D and IPY08 (sample numbers 76 and 18, respectively; Supplemental Table 2; posterior probability of 1.00). The estimated age of their hypothetical ancestor is between 203 and 4,439 years. These three individuals were grouped within the D1g haplogroup, whose node age was estimated to be between 12,555 and 17,713 years but with low posterior probability (0.653), a characteristic that was generally observed among sublineage relationships (Figure 2; Supplemental Figure 2).

The haplotype network also supports the inclusion of individual PZ3 within haplogroup D1g, with a derived haplotype from that of YA2D and IPY08; the three were distanced by at least six mutations from the haplogroup’s common ancestor (Figure 3). Moreover, this analysis reconstructs D1g sub-haplogroups with good support. The Bayesian skyline plot (BSP; Figure 4) shows an increase in population size between 10,000 and 15,000 years ago, overlapping with the age range obtained by Brandini and others (2017) for South American mitogenomes.

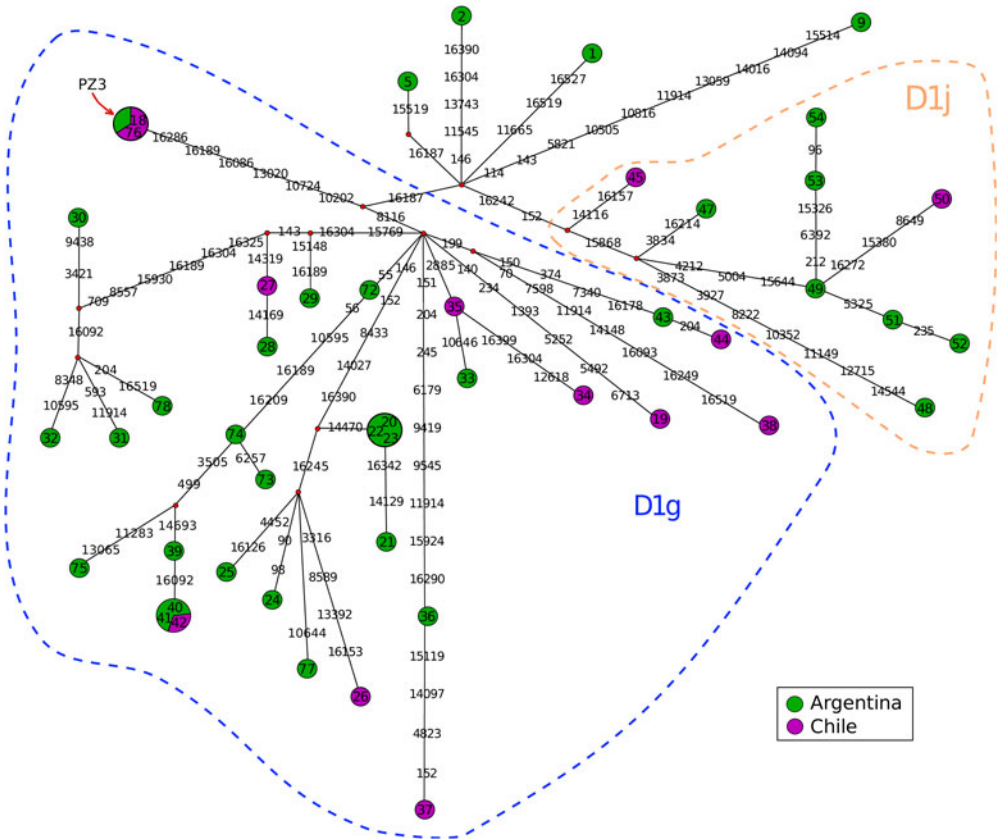


Figure 3. Haplotype median-joining network of the D1 lineage including individuals from Argentina and Chile constructed using Network (v10.2). Positions 309.1, 315.1, 16182, 16183, and 16193.1, as well as positions with gaps and missing values, were not considered.

Discussion

This article presents the first mitogenome entirely generated in Argentina, made possible by the pooled resources and joint efforts of researchers from several local institutions. The analysis performed did not find signs of contamination, and the mitogenome obtained presented the damage patterns expected for ancient DNA samples, although a complete profile could not be achieved due to the use of the USER enzyme during the library assembly. In addition, the pattern of mutation distribution (Table 1) was consistent with expectations for the mitogenome (Soares et al. 2009), attesting to the reliability of our results. The mitochondrial haplotype of individual PZ3 was assigned to the D1g haplogroup, one of the most frequently found in Patagonia (Bodner et al. 2012). In addition, the topology obtained by Bayesian phylogenetic reconstruction (Figure 2) supports this conclusion (posterior probability of 0.653). It also suggests that the PZ3 haplotype was closely related to a Yamana one—YA2D—from Navarino Island, as well as to an individual of unknown age, IPY08, from Hoste Island, located near the Seno Año Nuevo; both islands are in the Chilean Antarctic province (Figure 1).

Although with low posterior probability, the age of D1g node was estimated at between 12,555 and 17,713 years (Figure 2), concordant with the 20.9–11.7 thousands of years range estimated by Roca-Rada and colleagues (2021). It also partially overlaps with the $18,300 \pm 2400$ age range estimated for this clade by Bodner and colleagues (2012), who suggested that the D1g and D1j lineages could have been part of the genetic diversity of the first humans to settle in South America. Our estimate for the age of D1g is also consistent with the genetic and archaeological evidence of the earliest human occupation of this subcontinent (Brandini et al. 2017; Fagundes et al. 2008; Tamm et al. 2007). Moreover, the population increase observed between 10,000 and 15,000 years ago in the

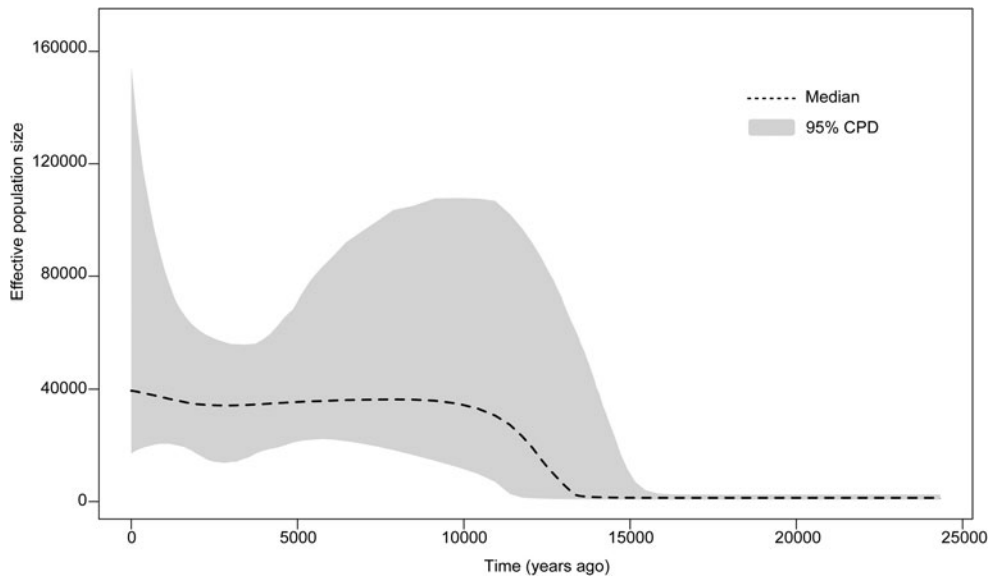


Figure 4. Bayesian skyline plot representing historical demographic trends in sampled D1 individuals. Time is reported on the x-axis as years BP, and the effective population size change is reported on the y-axis. Estimations were based on a mutation rate of 0.024694 substitutions per site per million years (Llamas et al. 2016), and y-axis transformation was performed considering a generational time of 25 years (as used in Brandini et al. 2017).

Bayesian skyline plot (Figure 4) is consistent with the age range for population growth estimated by Brandini and colleagues (2017) based on multiple South American mitogenomes. According to the authors, this expansion corresponds to the entry of human populations into South America.

From a more general perspective, it is worth noting that most of the D1g sub-haplogroups were reconstructed with posterior probability greater than 0.85 (Figure 2). However, relationships among these sub-haplogroups and between them and the PZ3-YA2D-IPY08 clade cannot be determined with certainty, given the low support of higher-level nodes (the deepest in time). This pattern can be observed throughout the entire phylogeny and can also be appreciated by qualitative analysis of the consensus trees obtained in this study (Supplemental Figure 2), where uncertainties regarding tree topology increase with the node level. This reveals a weak phylogenetic signal at the level of sub-haplogroup relationships, possibly due to the group's youth in evolutionary terms. Alternatively, the weak signal could be an artifact caused by poor sampling of the branches (only a few sequences are available for each sub-haplogroup). To our knowledge, there are no other published data regarding node support for this haplogroup. To provide more accurate answers regarding the population events that could be responsible for the observed pattern, it is necessary to pursue this line of inquiry and expand the database.

The age of the most recent common ancestor shared by PZ3, YA2D, and IPY08 was estimated at between 203 and 4,439 years (Figure 2). This wide age range is probably due to the paucity of information provided by the few and evolutionarily scarcely relevant differences between these three sequences. At that time, populations with maritime economies had already settled and stabilized in the southern Tierra del Fuego Archipelago, possibly as early as 6400 years BP (Orquera et al. 2011; Orquera and Piana 2009). The median haplotype network (Figure 3) showed that YA2D, IPY08, and PZ3 mitogenomes, the only insular individuals in our database, are separated from the hypothetical common ancestor shared with the rest of the D1g clade by at least six mutations: C10202T, T10724C, T13020C, T16086C, C16286T, and T16189C (the last one is considered to be a hotspot). The most noteworthy of the mutations are those in positions 16086 and 16286: they have also been reported in studies of the hypervariable region in both ancient and current individuals from the Beagle Channel area (Crespo et al. 2020; de la Fuente et al. 2015, 2018; de Saint Pierre et al. 2012;

Motti et al. 2020). In addition, the absence of the mutation in position 8116 is characteristic of Yaghan populations (de Saint Pierre et al. 2012).

These results may contribute to the discussion of at least two issues regarding population dynamics in the Tierra del Fuego archipelago. First, there are three types of archaeological evidence suggesting the existence of interactions between populations in the Beagle Channel and the Brunswick Peninsula-Seno Otway during the mid-Holocene (Figure 1): (1) the presence of green obsidian artifacts in Beagle Channel sites, a material that was frequently used in the Otway area archaeological sites (Álvarez 2004; Manzi 2004; Morello et al. 2015; Pallo 2016) and that would probably be obtained in raw form from somewhere in the Brunswick Peninsula (Emperaire and Laming-Emperaire 1961; Stern and Prieto 1991); (2) decoration designs and techniques found on similar artifact types in both areas (Fiore 2006); and (3) the distribution of subfoliar and lanceolate projectile points (Piana and Orquera 2007). However, the material evidence of this superregional interaction is no longer found in the archaeological record after about 5000–4000 years BP, suggesting spatial circumscription and a reduction in mobility due to several factors (Zangrando 2009). This change in the archaeological record coincides with our estimate for the maximum age of the most recent common ancestor of PZ3, YA2D, and IPY08. From this moment on, genetic drift may have fixated different lineages in the Beagle Channel and the islands of the western archipelago (de la Fuente et al. 2018). This process could also explain the preponderance of the D4h3a lineage over D1g and C1 lineages during the mid-Holocene in the western archipelago, an area associated with Alacalufe groups, and the dominance of C1 in the Beagle Channel population (de la Fuente et al. 2015; Moraga et al. 2010).

Second, the genetic proximity between PZ3 and IPY08 suggests the existence of gene flow or a recent common origin among the groups that occupied both sectors of the archipelago during the mid-Holocene. Although the sites where these individuals were recovered are only 90 km apart, there is very little information about the level of interaction between the Beagle Channel and southwestern areas of the archipelago during that period, mainly because there have been few archaeological explorations of Hoste Island. During historic times, Wedell (2006 [1825]:159–162) indicated the presence of different groups in the Seno Año Nuevo area, yet Bridges (1886) excluded the southern sector of Hoste Island from the Yaghan territory. Several authors have reported that the geographical distribution of Yaghan populations toward the western end of the archipelago was unclear during the nineteenth century: that area was also occupied by groups that could be ethnically and linguistically linked to Alacalufe populations (Orquera and Piana 1999). However, as has been suggested for the easternmost sector of the Fuegian archipelago (Crespo et al. 2020), the genetic diversity of populations before European contact indicates greater interaction between groups than what is suggested in historical documents. Even if we accept the existence of a genetic continuity between local populations pre- and post-European contact, the cultural geography of the channels and island south of Tierra del Fuego may have been highly dynamic.

Final Remarks

The mitogenome for individual PZ3 was obtained with an average depth of 8X and 99.7% coverage. The quality of the readings was high but there were few of them, which resulted in a relatively low average depth. Adjustments are being made to deal with these shortcomings. The haplotype was assigned to haplogroup D1g and was found to be closely related to YA2D and IPY08, a Yamana from Navarino Island and an ancient individual from Hoste Island. The estimated age for these mitogenomes' most recent common ancestor was 203–4,439 years (Figure 2), a time in which available archaeological evidence suggests that human groups with maritime economies were already settled in the southern Tierra del Fuego archipelago. The mitogenomes of PZ3, YA2D, and IPY08 present mutations that have only been described in past and extant Yaghan populations from the Beagle Channel area (Figure 3). The maximum age for the common ancestor coincides with a reduction in mobility in Beagle and the western archipelago islands at approximately 4500 years BP.

The construction of the molecular phylogeny made possible the reconstruction of most of D1g sub-haplogroups (Figure 2) and the estimation of age intervals for their nodes that were consistent with

results from previous studies. However, the relationships between these sub-haplogroups could not be determined with high certainty due to low support of the high-level nodes.

Finally, although massively parallel sequencing methodologies have been widely applied to aDNA studies, their implementation at the local level still poses many challenges. This first mitogenome entirely generated in Argentina faced some of these difficulties, but thanks to the collaboration of researchers from different local institutions it sets a solid precedent for future studies. This local workflow enabled local researchers to have greater control over sample processing and, in the future, will allow them to define and pursue locally relevant research goals and problems that are not necessarily aligned with research agendas in countries that traditionally led these kinds of studies.

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Data Availability Statement. The consensus mitogenome of individual PZ3 was deposited in GenBank under accession number ON422324. The raw data, in fastq format, are available at the European Nucleotide Archive under accession number PRJEB58193.

Competing Interests. The authors declare none.

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Supplemental Text 1. Materials and Methodology.

Supplemental Figure 1. Plots generated with MapDamage2 (v2.0; Jónsson et al. 2013) to visualize damage patterns. (A) Plots showing the base frequency outside and in the read (the open gray box corresponds to the read); (B) plots with the positions' specific substitutions from the 5' (left) and the 3' end (right). Color codes: red, C to T substitutions; blue, G to A substitutions; gray, all other substitutions; orange, soft-clipped bases; green, deletions relative to the reference; purple, insertions relative to the reference.

Supplemental Figure 2. DensiTree analyses showing the posterior distribution of the trees retrieved by BEAST. Dark regions indicate branch topology agreement among trees.

Supplemental Table 1. Illumina Adapters and Primers Used in this Article.

Supplemental Table 2. Database Compiled and Used in this Article. The information included regarding the haplogroup and the location or population of origin of the sample is the one provided by the paper that published the sequence (or its update in Phylotree17).

Supplemental Table 3. Sequence Data Statistics for the PZ3 Mitogenome.

Supplemental Table 4. Absolute Number of Changes between Haplotypes Performed in R (V3.4.4; R Core Team, 2020) with the Haplotypes Package (v1.1.2; Aktas 2020). Gaps were considered as a fifth character state. Accession numbers (or Sample ID when not available) and haplogroup are listed.

References Cited

- Aktas, Caner. 2020. Haplotypes: Manipulating DNA Sequences and Estimating Unambiguous Haplotype Network with Statistical Parsimony. R package version 1.1.2. <https://cran.r-project.org/web/packages/haplotypes/haplotypes.pdf>, accessed December 9, 2022.
- Álvarez, Myrian R. 2004. Estrategias tecnológicas en los grupos canoeros tempranos del área fuegopatagónica. *Magallania* 32:191–208.
- Andrews, Richard M., Iwona Kubacka, Patrick F. Chinnery, Robert N. Lightowlers, Douglass M. Turnbull, and Neil Howell. 1999. Reanalysis and Revision of the Cambridge Reference Sequence for Human Mitochondrial DNA. *Nature Genetics* 23:147.
- Aranda, Claudia, Gustavo Barrientos, and Mariano C. Del Papa. 2014. Código deontológico para el estudio, conservación y gestión de restos humanos de poblaciones del pasado. *Revista Argentina de Antropología Biológica* 16(2):111–113.
- Arencibia, Valeria, Cristian Crespo, Solana García Guraieb, María G. Russo, Cristina B. Dejean, and Rafael Goñi. 2019. Análisis genético poblacional de grupos cazadores recolectores del Holoceno tardío del Lago Salitroso (Santa Cruz, Argentina). *Revista Argentina de Antropología Biológica* 21(2):1–16.
- Asociación de Antropología Biológica Argentina. 2007. Declaración de la Asociación de Antropología Biológica Argentina en relación con la ética del estudio de restos humanos. <https://asociacionantropologiabiologicaargentina.org.ar>, accessed December 9, 2022.

- Barnett, Derek W., Erik K. Garrison, Aaron R. Quinlan, Michael P. Střmberg, and Gabor T. Marth. 2011. Bamtools: A C++ API and Toolkit for Analyzing and Managing BAM Files. *Bioinformatics* 27:1691–1692.
- Behar, Doron M., Mannis Van Oven, Saharon Rosset, Mait Metspalu, Eva Liis Loogvāli, Nuno M. Silva, Toomas Kivisild, Antonio Torroni, and Richard Villems. 2012. A “Copernican” Reassessment of the Human Mitochondrial DNA Tree from Its Root. *American Journal of Human Genetics* 90:675–684.
- Bodner, Martin, Ugo A. Perego, Gabriela Huber, Liane Fendt, Alexander W. Röck, Bettina Zimmermann, Anna Olivieri, et al. 2012. Rapid Coastal Spread of First Americans: Novel Insights from South America’s Southern Cone Mitochondrial Genomes. *Genome Research* 22:811–820.
- Bouckaert, Remco R., and Alexei J. Drummond. 2017. bModelTest: Bayesian Phylogenetic Site Model Averaging and Model Comparison. *BMC Evolutionary Biology* 17(1):42.
- Bouckaert, Remco, Timothy G. Vaughan, Joëlle Barido-Sottani, Sebastián Duchêne, Mathieu Fourment, Alexandra Gavryushkina, Joseph Heled, et al. 2019. BEAST 2.5: An Advanced Software Platform for Bayesian Evolutionary Analysis. *PLoS Computational Biology* 15(4):e1006650.
- Brandini, Stefania, Paola Bergamaschi, Marco Fernando Cerna, Francesca Gandini, Francesca Bastaroli, Emilie Bertolini, Cristina Cereda, et al. 2017. The Paleo-Indian Entry into South America according to Mitogenomes. *Molecular Biology and Evolution* 35:299–311.
- Bridges, Thomas. 1886. El confin de la República: La Tierra del Fuego y sus habitantes. *Boletín del Instituto Geográfico Argentino* 7:313–320.
- Briggs, Adrian W., Udo Stenzel, Philip L. F. Johnson, Richard E. Green, Janet Kelso, Kay Prüfer, Matthias Meyer, et al. 2007. Patterns of Damage in Genomic DNA Sequences from a Neandertal. *PNAS* 104:14616–14621.
- Cardozo, Darío Gonzalo, Cristina Beatriz Dejean, Gabriela Russo, Bárbara Mazza, Daniel Loponte, Alejandro Acosta, María Rosario Feuillet, and Silvia Corne. 2021. Impacto de la conquista hispánica en el humedal del Paraná inferior (Argentina): Una perspectiva histórica y genética mitocondrial. *Runa* 42:409–433.
- Crespo, Cristian M., Darío G. Cardozo, Augusto Tessone, Martín Vázquez, Caroline Kisielinski, Valeria Arencibia, Justin Tackney, A. Francisco Zangrando, and Cristina B. Dejean. 2020. Distribution of Maternal Lineages in Hunter-Gatherer Societies of the Southern Coast of Tierra Del Fuego, Argentina. *American Journal of Physical Anthropology* 173: 709–720
- Crespo, Cristian M., Cristian Favier Dubois, M. Gabriela Russo, José L. Lanata, and Cristina B. Dejean. 2017a. First Analysis of Ancient MtDNA Genetic Diversity in Northern Coast of Argentinean Patagonia. *Journal of Archaeological Science: Reports* 12:91–98.
- Crespo, Cristian M., José L. Lanata, Darío G. Cardozo, Sergio Avena, and Cristina B. Dejean. 2018. Ancient Maternal Lineages in Hunter-Gatherer Groups of Argentinean Patagonia. Settlement, Population Continuity and Divergence. *Journal of Archaeological Science: Reports* 18:689–695.
- Crespo, Cristian M., María Russo, Adam Hajduk, José Lanata, and Cristina Dejean. 2017b. Variabilidad mitocondrial en muestras pre-colombinas de la patagonia argentina: hacia una visión de su poblamiento desde el ADN antiguo. *Revista Argentina de Antropología Biológica* 19(1):1–21.
- Danecek, Petr, and Shane A. McCarthy. 2017. BCFtools/Csq: Haplotype-Aware Variant Consequences. *Bioinformatics* 33:2037–2039.
- De la Fuente, Constanza, María C. Ávila-Arcos, Jacqueline Galimany, Meredith L. Carpenter, Julian R. Homburger, Alejandro Blanco, Paloma Contreras, et al. 2018. Genomic Insights into the Origin and Diversification of Late Maritime Hunter-Gatherers from the Chilean Patagonia. *PNAS* 115:E4006–4012.
- De la Fuente, Constanza, Jacqueline Galimany, Brian M. Kemp, Kathleen Judd, Omar Reyes, and Mauricio Moraga. 2015. Ancient Marine Hunter-Gatherers from Patagonia and Tierra Del Fuego: Diversity and Differentiation Using Uniparentally Inherited Genetic Markers. *American Journal of Physical Anthropology* 158:719–729.
- De Saint Pierre, Michelle, Claudio M. Bravi, Josefina M. B. Motti, Noriyuki Fuku, Masashi Tanaka, Elena Llop, Sandro L. Bonatto, and Mauricio Moraga. 2012. An Alternative Model for the Early Peopling of Southern South America Revealed by Analyses of Three Mitochondrial DNA Haplogroups. *PLoS ONE* 7(9):e4348.
- Drummond, Alexei J., and Remco R. Bouckaert. 2015. *Bayesian Evolutionary Analysis with BEAST*. Cambridge University Press, Cambridge.
- Empereire, José, and Annette Laming-Empereire. 1961. Les gisements des îles Englefield et Vivian dans la mer d’Otway (Patagonie australe). *Journal de la Société des Américanistes* 50:7–77.
- Fagundes, Nelson J. R., Ricardo Kanitz, Roberta Eckert, Ana C. S. Valls, Mauricio R. Bogo, Francisco M. Salzano, David Glenn Smith, et al. 2008. Mitochondrial Population Genomics Supports a Single Pre-Clovis Origin with a Coastal Route for the Peopling of the Americas. *American Journal of Human Genetics* 82:583–592.
- Fehren-Schmitz, Lars, and Lea Georges. 2016. Ancient DNA Reveals Selection Acting on Genes Associated with Hypoxia Response in Pre-Columbian Peruvian Highlanders in the Last 8,500 Years. *Scientific Reports* 6:23485
- Fiore, Dánae. 2006. Puentes de agua para el arte mobiliario: La distribución espacio-temporal de artefactos óseos decorados en Patagonia meridional y Tierra del Fuego. *Cazadores-Recolectores del Cono Sur* 1:137–147.
- Jónsson, Hákon, Aurélien Ginolhac, Mikkel Schubert, Philip L. Johnson, and Ludovic Orlando. 2013. MapDamage2.0: Fast Approximate Bayesian Estimates of Ancient DNA Damage Parameters. *Bioinformatics* 29:1682–1684.
- Jun, Goo, Mary Kate Wing, Gonçalo R. Abecasis, and Hyun Min Kang. 2015. An Efficient and Scalable Analysis Framework for Variant Extraction and Refinement from Population-Scale DNA Sequence Data. *Genome Research* 25:918–925.
- Katoh, Kazutaka, Kazuharu Misawa, Kei-ichi Kuma, and Takashi Miyata. 2002. MAFFT: A Novel Method for Rapid Multiple Sequence Alignment Based on Fast Fourier Transform. *Nucleic Acids Research* 30:3059–3066.

- Kreimer, Pablo. 2006. ¿Dependientes o integrados? La ciencia latinoamericana. *Nómadas* 24:199–212.
- Kreimer, Pablo, and Victoria Ugartemendía. 2007. Ciencia en la universidad: Dimensiones locales e internacionales. *Atos de Pesquisa Em Educação* 2:461–485.
- Lalueza, Carles, Alejandro Pérez-Pérez, Eva Prats, Lluís Cornudella, and Daniel Turbón. 1997. Lack of Founding Amerindian Mitochondrial DNA Lineages in Extinct Aborigines from Tierra del Fuego–Patagonia. *Human Molecular Genetics* 6:41–46.
- Lalueza Fox, Carles. 1996. Mitochondrial DNA Haplogroups in Four Tribes from Tierra del Fuego—Patagonia: Inferences about the Peopling of the Americas. *Human Biology* 68:855–871.
- Li, Heng, and Richard Durbin. 2009. Fast and Accurate Short Read Alignment with Burrows–Wheeler Transform. *Bioinformatics* 25:1754–1760.
- Lindo, John, Randall Haas, Courtney Hofman, Mario Apata, Mauricio Moraga, Ricardo A. Verdugo, James T. Watson, et al. 2018. The Genetic Prehistory of the Andean Highlands 7000 Years BP through European Contact. *Science Advances* 4:eau4921.
- Llamas, Bastien, Lars Fehren-Schmitz, Guido Valverde, Julien Soubrier, Swapan Mallick, Nadin Rohland, Susanne Nordenfelt, et al. 2016. Ancient Mitochondrial DNA Provides High-Resolution Time Scale of the Peopling of the Americas. *Science Advances* 2:e1501385.
- Manzi, Liliana M. 2004. La obsidiana verde como indicador de formas de uso del espacio en fuego-patagonia. In *Contra viento y marea: Arqueología de Patagonia*, edited by María Teresa Civalero, Pablo Fernández, and Gabriela Guráieb, pp. 117–134. Instituto Nacional de Antropología y Pensamiento Latinoamericano, Buenos Aires.
- Marcic, Tomislav, Mark Whitten, and Svante Pääbo. 2010. Multiplexed DNA Sequence Capture of Mitochondrial Genomes Using PCR Products. *PLoS ONE* 5(11):e14004.
- Martin, Marcel. 2011. Cutadapt Removes Adapter Sequences from High-Throughput Sequencing Reads. *EMBnet.Journal* 17(1):10–12.
- Maturana Russel, Patricio, Brendon J. Brewer, Steffen Klaere, and Remco R. Bouckaert. 2019. Model Selection and Parameter Inference in Phylogenetics Using Nested Sampling. *Systematic Biology* 68:219–233.
- Moraga, Mauricio, Michelle de Saint Pierre, Francisco Torres, and Juan Ríos. 2010. Kinship by Maternal via Between the Last Descendants of Kaweskar Ethnicity and Burials in the Patagonian Channels: Evidence from the Study of Mitochondrial Lineages. *Magallania* 38(2):103–114.
- Morello, Flavia, Charles Stern, and Manuel San Román. 2015. Obsidiana verde en Tierra del Fuego y Patagonia: Caracterización, distribución y problemáticas culturales a lo largo del Holoceno. *Intersecciones en Antropología* 16(1):139–153.
- Motti, Josefina M. B., Erika Hagelberg, John Lindo, Ripan S. Malhi, Claudio M. Bravi, and Ricardo A. Guichón. 2015. Primer genoma mitocondrial en restos humanos de la costa de Santa Cruz, Argentina. *Magallania* 43(2):119–131.
- Motti, Josefina M. B., A. Sebastián Muñoz, Isabel Cruz, Manuel D. D'Angelo del Campo, Luis A. Borrero, Claudio M. Bravi, and Ricardo A. Guichón. 2019. Análisis de ADN mitocondrial en restos humanos del Holoceno tardío del sur de Santa Cruz. In *Arqueología de la Patagonia: El pasado en las arenas*, edited by Julieta Gómez Otero, Ariadna Svoboda, and Anahí Banegas, pp. 493–503. Altuna Impresores, Buenos Aires.
- Motti, Josefina M. B., Stevie Winingear, L. O. Valenzuela, M. A. Nieves-Colón, Kelly M. Harkins, P. García Laborde, C. M. Bravi, R. A. Guichón, and A. C. Stone. 2020. Identification of the Geographic Origins of People Buried in the Cemetery of the Salesian Mission of Tierra del Fuego through the Analyses of mtDNA and Stable Isotopes. *Journal of Archaeological Science: Reports* 33:102559.
- Nakatsuka, Nathan, Pierre Luisi, Josefina M. B. Motti, Mónica Salemme, Fernando Santiago, Manuel D. D'Angelo del Campo, Rodrigo J. Vecchi, et al. 2020. Ancient Genomes in South Patagonia Reveal Population Movements Associated with Technological Shifts and Geography. *Nature Communications* 11:3868.
- Narasimhan, Vagheesh, Petr Danecek, Aylwyn Scally, Yali Xue, Chris Tyler-Smith, and Richard Durbin. 2016. BCFTools/RoH: A Hidden Markov Model Approach for Detecting Autozygosity from Next-Generation Sequencing Data. *Bioinformatics* 32:1749–1751.
- Orquera, Luis A., Dominique Legoupil, and Ernesto L. Piana. 2011. Littoral Adaptation at the Southern End of South America. *Quaternary International* 239:61–69.
- Orquera, Luis A., and Ernesto L. Piana. 1999. *Arqueología de la región del Canal Beagle (Tierra del Fuego, República Argentina)*. Sociedad Argentina de Antropología, Buenos Aires.
- Orquera, Luis A., and Ernesto L. Piana. 2009. Sea Nomads of the Beagle Channel in Southernmost South America: Over Six Thousand Years of Coastal Adaptation and Stability. *Journal of Island and Coastal Archaeology* 4:61–81.
- Pallo, María Cecilia. 2016. Obsidian Transport in Fuego-Patagonia (Southernmost South America): A Spatial Statistical Approach. *Journal of Anthropological Archaeology* 44:114–126.
- Parolin, María Laura, Jacqueline Galimany, Julieta Gómez Otero, Silvia L. Dahinten, A. Gabriela Millán, and Mauricio Moraga. 2019. Primeras secuencias mitocondriales de la región control completa en muestras humanas del Holoceno tardío de la costa norte y centro de Patagonia, Argentina. In *Arqueología de la Patagonia: El pasado en las arenas*, edited by Julieta Gómez Otero, Ariadna Svoboda, and Anahí Banegas, pp. 469–480. Altuna Impresores, Buenos Aires.
- Perez, S. Ivan, María Bárbara Postillone, Diego Rindel, Diego Gobbo, Paula N. Gonzalez, and Valeria Bernal. 2016. Peopling Time, Spatial Occupation and Demography of Late Pleistocene–Holocene Human Population from Patagonia. *Quaternary International* 425:214–223.
- Piana, Ernesto L., and Luis A. Orquera. 2007. Diferencias regionales y temporales en el litoral sur sudamericano. In *Arqueología de Fuego-Patagonia: Levantando piedras, desenterrando huesos . . . y develando arcanos*, edited by Flavia Morello, Mateo Martinic, Alfredo Prieto, and Gabriel Bahamonde, pp. 311–326. Ediciones CEQUA, Punta Arenas, Chile.
- Posth, Cosimo, Nathan Nakatsuka, Iosif Lazaridis, Pontus Skoglund, Swapan Mallick, Thiseas C. Lamnidis, Nadin Rohland, et al. 2018. Reconstructing the Deep Population History of Central and South America. *Cell* 175:1185–1197.

- Postillone, María Bárbara, Gustavo Martínez, Gustavo Flensburg, and Cristina B. Dejean. 2020. First Analysis of Mitochondrial Lineages from the Eastern Pampa–Patagonia Transition during the Final Late Holocene. *American Journal of Physical Anthropology* 171:659–670.
- Raghavan, Maanasa, Matthias Steinrücken, Kelley Harris, Stephan Schiffels, Simon Rasmussen, Michael DeGiorgio, Anders Albrechtsen, et al. 2015. Genomic Evidence for the Pleistocene and Recent Population History of Native Americans. *Science* 349:aab3884.
- Rambaut, Andrew. 2018. FigTree version 1.4.4: Tree Drawing Tool in Computer Program and Documentation. <http://tree.bio.ed.ac.uk/software/figtree/>, accessed December 9, 2022.
- Rambaut, Andrew, Alexei J. Drummond, Dong Xie, Guy Baele, and Marc A. Suchard. 2018. Posterior Summarization in Bayesian Phylogenetics Using Tracer 1.7. *Systematic Biology* 67:901–904.
- R Development Core Team. 2020. R: A Language and Environment for Statistical Computing. Foundation for Statistical Computing. <https://softlibre.unizar.es/manuales/aplicaciones/r/fullrefman.pdf>, accessed December 9, 2022.
- Roca-Rada, Xavier, Gustavo Politis, Pablo G. Messineo, Nahuel Scheifler, Clara Scabuzzo, Mariela González, Kelly M. Harkins, et al. 2021. Ancient Mitochondrial Genomes from the Argentinian Pampas Inform the Early Peopling of the Southern Cone of South America. *IScience* 24:102553.
- Russo, M. Gabriela, Cristina B. Dejean, Sergio A. Avena, Verónica Seldes, and Paola Ramundo. 2018. Mitochondrial Lineage A2ah Found in a Pre-Hispanic Individual from the Andean Region. *American Journal of Human Biology* 30:e23134.
- Russo, M. Gabriela, Fanny Mendisco, Sergio A. Avena, Cristina B. Dejean, and Verónica Seldes. 2016. Pre-Hispanic Mortuary Practices in Quebrada de Humahuaca (North-Western Argentina): Genetic Relatedness among Individuals Buried in the Same Grave. *Annals of Human Genetics* 80: 210–220.
- Sawyer, Susanna, Johannes Krause, Katerina Guschanski, Vincent Savolainen, and Svante Pääbo. 2012. Temporal Patterns of Nucleotide Misincorporations and DNA Fragmentation in Ancient DNA. *PLoS ONE* 7(3):e34131.
- Schubert, Mikkel, Stinus Lindgreen, and Ludovic Orlando. 2016. AdapterRemoval v2: Rapid Adapter Trimming, Identification, and Read Merging. *BMC Research Notes* 9:88.
- Soares, Pedro, Luca Ermini, Noel Thomson, Maru Mormina, Teresa Rito, Arne Röhl, Antonio Salas, Stephen Oppenheimer, Vincent Macaulay, and Martin B. Richards. 2009. Correcting for Purifying Selection: An Improved Human Mitochondrial Molecular Clock. *American Journal of Human Genetics* 84:740–759.
- Stern, Charles, and Alfredo Prieto. 1991. Obsidiana verde de los sitios arqueológicos en los alrededores del seno Otway, Magallanes, Chile. *Anales del Instituto de la Patagonia* 20:139–144.
- Suby, Jorge A., Atilio F. Zangrando, and Ernesto Piana. 2011. Exploraciones osteológicas de la salud de las poblaciones humanas del Canal Beagle. *Relaciones de la Sociedad Argentina de Antropología* 36:249–270.
- Tamm, Erika, Toomas Kivisild, Maere Reidla, Mait Metspalu, David Glenn Smith, Connie J. Mulligan, Claudio M. Bravi, et al. 2007. Beringian Standstill and Spread of Native American Founders. *PLoS ONE* 2(9):e829.
- Tavella, María Pía, Darío A. Demarchi, and Rodrigo Nores. 2020. Diversidad genética en restos humanos de contextos arqueológicos tardíos de la provincia de Santiago del Estero a partir del estudio de ADN mitocondrial. *Revista Argentina de Antropología Biológica* 22:013.
- Van Oven, Mannis, and Manfred Kayser. 2009. Updated Comprehensive Phylogenetic Tree of Global Human Mitochondrial DNA Variation. *Human Mutation* 30:E386–394.
- Wade, Lizzie. 2018. Ancient DNA Tracks Migrations around Americas. *Science* 362:627–628.
- Wedell, James. 2006 [1825]. *Un viaje hacia el Polo Sur realizado en los años 1822–1824*. Eudeba, Universidad de Buenos Aires.
- Weissensteiner Hansi, Dominic Pacher, Anita Kloss-Brandstätter, Lukas Forer, Günther Specht, Hans-Jürgen Bandelt, Florian Kronenberg, Antonio Salas, and Sebastian Schönherr. 2016. HaploGrep 2: Mitochondrial Haplogroup Classification in the Era of High-Throughput Sequencing. *Nucleic Acids Research* 44:W58–W63.
- Zangrando, Atilio F. 2009. *Historia evolutiva y subsistencia de cazadores-recolectores marítimos de Tierra del Fuego*. Colección Tesis de Doctorado, Sociedad Argentina de Antropología, Buenos Aires.
- Zangrando, Atilio F., Hein B. Bjerck, Ernesto L. Piana, Heidi M. Breivik, Angélica M. Tivoli, and Joan Negre. 2018. Spatial Pattern and Occupation Dynamics during the Early Holocene in an Archaeological Site from the South Coast of Tierra del Fuego: Binushmuka I. *Estudios Atacameños* 60:31–49.
- Zangrando, Atilio F., Angélica M. Tivoli, Juan F. Ponce, Daniela V. Alunni, María del Carmen Fernández Roper, and María Paz Martinoli. 2022. Colonización humana de Tierra del Fuego: Contextos arqueológicos pre-transgresivos en la costa norte del Canal Beagle. *Chungara* 54:557–577.

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