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MOLECULAR APPROACH TO THE PEOPLING OF THE AMERICAS BY SEQUENCING mtDNA FROM EXTINCT FUEGUIANS AND PATAGONS

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The study of the aborigines of Tierra del Fuego-Patagonia is of particular interest for reconstructing the first population of America. The ancestors of the Aonikenk, Selknam, Yamana and Kaweskar ethnic groups, today extinct, settled in the extreme south of America about 12,000 years ago as the region thawed. Some of these groups have lived in considerable isolation since then. Therefore the genetic information, interesting from a phylogenetic point of view, that can be obtained from ancient DNA will reflect the peculiarities of those descendants of the first Americans. Ancient DNA studies of extinct groups make it possible to complete or confirm those carried out on current populations. In the case of the American extreme South, other Amerinds migrated to the zone in recent times mainly to exploit economic resources. The degree to which they intermixed with each other and with the Fueguians that survived extinction, if any did, is unknown.

The development of the PCR technique has greatly facilitated the analysis of the genetic material of our ancient samples. The current study analyses the conclusions of an analysis of nineteen sequences from the Hypervariable Region of mtDNA Control obtained from our ancient samples (between 4,000 and 200 years a.P.), mainly dental pieces, of extinct individuals of the different Fuegian and Patagonian aborigines. This genetic marker is very useful for our purpose because it is inherited exclusively from the mother, the absence of recombination and its high mutation rate, which generates the necessary variability for distinguishing populations.

The DNA is extracted by the usual molecular biology procedures with a few particularities. The samples were crushed in a liquid nitrogen refrigerated grinder, washed with an EDTA 0.5 M and digested with proteinase K. Then the DNA was extracted with Phenol/Chloroform and was concentrated with Centricon-30 microconcentrators. Finally fragments corresponding to the mtDNA Hypervariable Region I were amplified and sequenced with specific primers yielding a PCR product of 273 pairs of bases.

Previous studies have identified four main mitochondrial haplogroups among Amerinds, called A, B, C and D. Some of the differences observed with respect to the referred sequence (Anderson) correspond to mutations that are characteristic of haplogroup C and D, the only two observed among the aborigines of Tierra del Fuego and Patagonia. The analysis of variability of the obtained sequences was performed within the context of the current Amerind populations published by other authors. The genetic dissimilarity between the sequences was calculated by the Kimura-2 distance parameters and, then, generating affinity trees by means of neighbour-joining and UPGMA methods respectively (not shown).

An extremely meticulous previous study indicated, after considering almost 500 trees, that the C and D individuals studied here were separated in 97% of all the trees studied. This confers a robust basis to the separation of the two haplogroups and the groupings observed among the Fueguians and the Patagons by means of neighbor-joining and UPGMA.

The sequences of the Fueguian and Patagonian aborigines obtained cluster in haplogroups C and D, which confirms the results obtained previously with RFLPs ([Lalueza et al. 1997](#)). This lends reliability to the sequences obtained.

Some of the sequences obtained are located at the roots of the trees and others integrate with the set of current Amerindians. In our opinion, the samples that stand away from the main set of each haplogroup would correspond to descendants of the first inhabitants of the American Continent because of the differences they present with respect to the rest of the Amerindians. This is coherent with the fact that some Asiatics and Amerindians (Han from Taiwan, Koreans and Apaches, for example) group among themselves and far from Fueguians and Patagons. This may indicate that the inhabitants of Tierra del Fuego-Patagonia would be descendants of an initial wave of migration towards America different from other later migratory episodes (see [Lahr 1995](#)). Later, the demographic and geographic expansions of the American Neolithic could have absorbed the minority population groups. This is supported by morphology studies ([Lahr 1995](#)) and difficult to detect from the single origin hypothesis of [Merriwether et al. \(1995\)](#).

The presence of some sequences that also stand away from the main set of each haplogroup of Amerindians from Central and North America could represent, in turn, mitochondrial descendants of the ancient migrations absorbed by later migratory episodes. However, the possibility that these cases are retro-mutations should be considered, since they are not very numerous.

On the other hand, some Fueguian sequences fall well within the general Amerind variability and this could be interpreted as corresponding to later accretions arriving from Tierra del Fuego-Patagonia. Future studies, with more information, will contribute to clarifying whether or not hypothesis proffered here can be confirmed.

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