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Megaplex analysis of a Mongolian population from the Egyin Gol site (300 B.C.–300 A.D.)

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Abstract

Genetic analysis of archaeological remains may provide information of prime importance to the understanding of human past history. In order to investigate the history of Mongolian populations, we have analyzed the skeletal remains of 56 specimens excavated from the Egyin Gol necropolis located in northern Mongolia. This burial site is linked to the Xiongnu period, which extended from the 3rd century B.C. to the 3rd century A.D. The environmental conditions at this site were cold and dry, and the DNA preservation was exceptional, allowing nuclear markers such as microsatellites to be studied. Molecular analyses were performed on long cortical bone samples with the AmplF*l*STR Profiler Plus kit (Applied Biosystems). Amplifications were successful for 47 of the 56 specimens analysed. Molecular sexing confirmed archeological data based on morphometric parameters and allowed the gender of four juvenile skeletons to be determined. Short tandem repeat (STR) analysis showed close relationships between several specimens and provides additional background information concerning social organization within the necropolis as well as funeral practices linked to Xiongnu populations.

Keywords: Ancient DNA; Short tandem repeat; Genetic analysis; Xiongnu; Mongolian population

1. Introduction

In recent years, molecular studies have become widely employed as a tool for investigating parentage relationships within burial groups [1,2]. Indeed, the knowledge

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of genetic relationships within and between burial sites allows a better understanding of the organization of the sepulchral places and of the origin of the human remains recovered [3].

In the present study, we examined biological kinship in a necropolis from the Xiongnu period (400 B.C.–300 A.D.). The Xiongnu were ancient nomadic people who represented an organized threatening force; their repeated invasions prompted the Empire of China to begin erecting what later became the Great Wall.

The site, located near the Egyin Gol river in Northern Mongolia, was erected between the 3rd century B.C. to the 3rd century A.D. The climatic conditions encountered at this site had protected the recovered specimens against DNA degradation, allowing nuclear markers such as short tandem repeats (STRs) to be studied. STRs represent propitious markers for ancient DNA studies due to their high discriminatory power, their small size, the possibility to amplify several of them simultaneously, and finally their ability to give an indication for the authenticity of results.

2. Materials and methods

Molecular analyses were performed in most cases on long cortical bones. To eliminate surface contamination, the outer surface of the bones was removed to almost 3 mm in depth with the use of a sanding machine. Powdered bone was then generated with a drill equipped with a surgical trepan. DNA was extracted according to a published protocol [4].

Autosomal STR amplifications were performed using the AmpF*l*STR profiler Plus™ Kit (Applied Biosystems) which allows the amplification of nine STRs loci and the amelogenin locus (determining the individual's sex).

To ensure the accuracy and reliability of the results, all samples were amplified at least six times from two independent extracts. Moreover, the extensive precautions imposed when working on ancient DNA [5] have been taken during all steps of the analysis.

3. Results

Amplifications were successful for 47 of the 56 specimens analysed. Four samples failed to yield any detectable human nuclear DNA in several independent extracts, whereas five contained too few template molecules to provide reproducible results. The remaining samples gave more or less complete allelic profiles.

Molecular sexing confirmed archeological data based on morphometric parameters. In addition, it allowed the gender of four juvenile skeletons to be determined where the morphological indicators of sex were absent or uninformative.

Comparison of the profiles in pairs allowed the identification of a family composed of the father, the mother and one child (Fig. 1, samples 57, 59, and 58). It was also possible to determine other familial relationships indicated by arrows on the map of the necropolis (Fig. 1). Statistic analysis using the Genetix software revealed that each of the three sectors

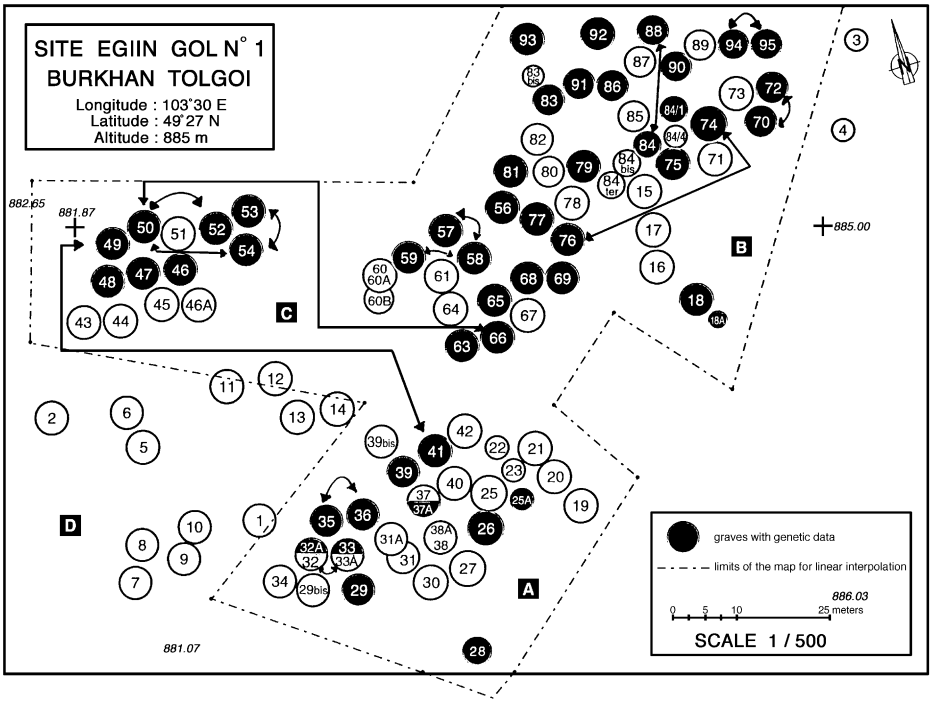

Fig. 1. Map of the necropolis. Graves are represented by circles. Those for which genetic analyses have been made are represented by dark circles. Letters A, B, C and D represent the four sectors that had been distinguished. Dotted lines define the boundary of these sectors. Close genetic relationships are indicated by arrows.

A, B, and C, defined by the anthropologists apparently corresponds to groups of genetically linked individuals.

4. Discussion

The present study shows the importance of nuclear genetic analyses performed in burial sites to facilitate the interpretation of the social organization within the necropolis, as well as to highlight some funeral practices linked to ancient populations. From our results it was possible to deduce that the implementation of the graves in the Egyin Gol necropolis may have responded to several criteria: (1) the presence of relatives (except for two cases, genetically related individuals were found in graves close to each other), (2) the individuals belonged to a family group (A, B, C) and (3) the apparent affiliation to a social elite. Indeed, the small number of inhumations that occurred throughout more than four centuries suggested that this Xiongnu tribe probably had other sepulchral places.

In the future, other approaches such as Y chromosomal STR typing or analysis of the hypervariable regions of the mitochondrial DNA will be tested to further understand the history of this necropolis.

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