Got her mummy´s eyes
Eye color investigation using IrisPlex – preliminary study

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Introduction

SNP phenotypic markers are being studied by several groups worldwide. As a result, phenotypic loci such as the ones responsible for human eye, hair and skin color are starting to get known. These may provide information for identification of unknown sample donors in criminal casework when there are no suspects. This kind of information may be even more relevant when the sample donor possesses phenotypic characteristics which distinguish him from the population in which he is inserted. Nevertheless, this approach may be used in similar scientific areas.

In this work, a DNA sample from a mummified corpse with historical and scientific interest was studied in order to discover more information about who this corps belonged to. One of the tools used in this investigation was IrisPlex, designed for eye color prediction. This study presents one example on how this methodologies may be useful, not only in forensic investigation but also in areas such as physical anthropology.

Results and Discussion

➢ The obtained results from DNA quantification and from DNA genotyping with Promega Powerplex® 16 HS System, revealed that the cadaverous remains belonged to a female individual, and that she probably belonged to Eurasia population group. Based on information found on the corpse, it is believed that this individual was deceased in the years 30-40 of the last century (data not shown).

➢ Based on the profile obtained with IrisPlex phenotyping of the mummy's sample (Figure 1B - 1G / A, 2C, 3G / T, 4G, 5G, 6C), it was estimated that the individual would have a 71% probability of having brown eyes (Table 1). The model used for this estimate has an accuracy of 92.5% for the class of brown eyes.

➢ Using for comparison a female individual with the same phenotypic profile as genotyped in the mummy, we perceive that the mummified individual could had have brown eyes similar to the ones of that women (Figure 1 and Table 1).

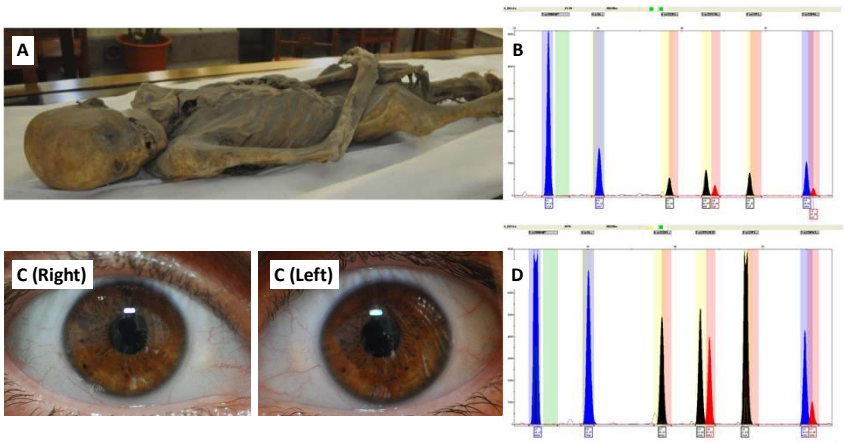

Figure 1 – (A) Mummified corpse belonging to INMLCF on which there is no information about its identity, and (B) respective IrisPlex phenotypic profile. (C) Brown eyes that exemplify what probably was the eye color of the mummy in study, since they correspond to a (D) female individual who has the same phenotypic profile.

Table 1 – Probabilities for eye color of the referred individuals (mummified corpse and female individual used for exemplification).

	P. blue	P. intermediate	P. brown
Mummified corpse (♀)	15 %	14 %	71 %
Female sample (♀)	15 %	14 %	71 %

Material and Methods

✓ A muscle fragment from the knee joint region was collected from a mummified corpse (Figure 1);
✓ DNA was extracted with *QIAamp DNA Mini kit (Qiagen)* and quantified with *Quantifiler® Duo* in *Applied Biosystems 7500 Real-Time PCR System*;
✓ STRs were genotyped with *Powerplex® 16 HS kit (Promega)* and SNPs were phenotyped with IrisPlex (loci HERC2 rs12913832, OCA2 rs1800407, SLC24A4 rs12896399, SLC45A2 rs16891982, TYR rs1393350, and IRF4 rs12203592). For both type of markers, PCR amplification was realized in *Applied Biosystems GeneAmp® PCR System 9700* thermocycler and subsequent capillary electrophoresis and analysis performed in *Applied Biosystems 3130 system*;
✓ Population group affiliation analysis was realized using *PopAffiliator* v 1.1 e 2.1 software;
✓ Estimates used for the analysis of eye color were based on the model of Liu et al. Phenotypic profile of mummy was compared to profiles obtained in project currently running.

Conclusions

Phenotypic information about a mummified individual was obtained, presumably a corpse from years 1930-1940. From the sample, which was relatively old and degraded, was possible to determine with high probability what was the individual's eye color.

Although the presented results have more historical and scientific than forensic interest, this study aims to illustrate that phenotypic markers may serve not only as a tool in criminal investigation but also as an instrument in the anthropology field. Concluding, phenotypic markers may provide additional information when the study of cadaveric remains of forensic or historical interest is necessary.

Bibliography

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