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# EVIDENCE FOR AN ANCIENT ORIGIN OF THE FGA p.Glu545Val (E526V) AMYLOIDOSIS-CAUSING MUTATION ENDEMIC IN NORTHERN PORTUGAL

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## Background

Fibrinogen alpha chain amyloidosis is widely spread throughout the world, and is most frequently associated with the *FGA* p.Glu545Val (E526V) mutation, particularly in European countries, with endemic foci of the disease identified in the UK, and in northern Portugal (1) (Figure 1). A preliminary attempt to characterize the disease-associated haplotype hinted at a common ancestor (2), but whether this is true and how far back in time the founding event would have taken place is still much an open question.

## Study Cohort

All available, previously characterized Portuguese patients and relatives: 56 individuals in total, 33 of which were disease mutation carriers, belonging to 12 extended families.

A control population of 67 unrelated individuals from northern Portugal was used.

## Methods

Thirteen polymorphic short tandem-repeats (STRs) spanning 5.8Mbps over the *FGA* gene in chromosome 4, from the initial set of 19 selected, were successfully genotyped using a PCR multiplex assay and custom designed oligonucleotides. Assay products were separated in an Applied Biosystems genetic analyzer and sized using the microsatellite assay module provided on the Thermo Fisher Connect cloud.

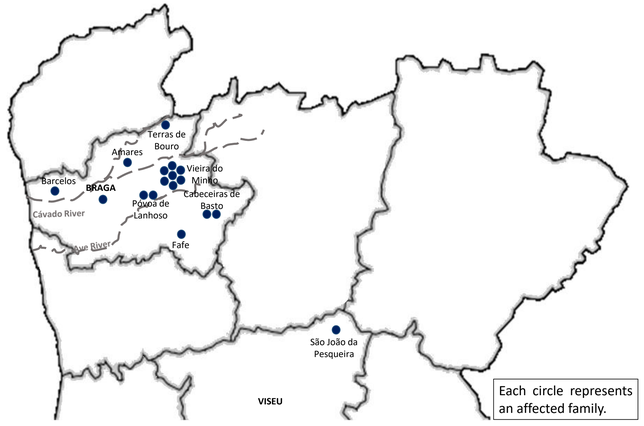
Haplotype phasing was carried out using an empirical linkage disequilibrium-based method, implemented in the Beagle 4.0 computer program, release 1399 (3) with some adjustments to accommodate pedigree constraints and to preserve parsimony. The age of the E526V mutation in this population was calculated by Bayesian estimation of a multipoint LD model, as implemented in the DMLE+ program, v. 2.3 (4).

## Results

Seven different but closely related disease-associated haplotypes were identified, the most frequent of which, represented in 5 families, was presumed to be the ancestral haplotype (Table 1).

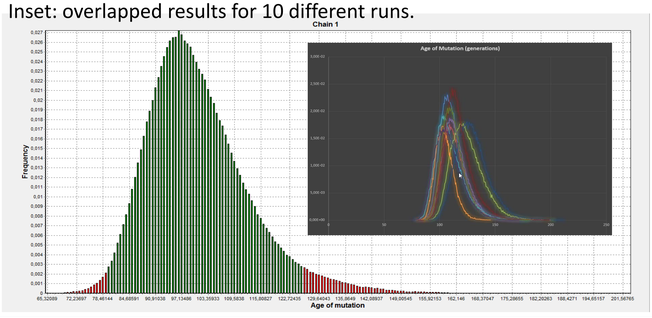
The linkage disequilibrium-based model implemented in DMLE+, although somewhat sensitive to estimates of population growth rate and other parameters, consistently predicted a mutation age exceeding 100 generations (2500 years) (Figure 2).

### Figure 1. Geographic distribution of families with AFibE526V (p.Glu545Val) amyloidosis in Portugal




### Figure 2. LD Model fitting (DLME+)

Representative posterior probability distribution for age of mutation.
Inset: overlapped results for 10 different runs.




## References

1. Tavares I, Oliveira JP, Pinho A, et al. Unrecognized Fibrinogen A α-Chain Amyloidosis: Results From Targeted Genetic Testing. *Am J Kidney Dis*. 2017;70(2):235-243. doi:10.1053/j.ajkd.2017.01.048
2. Tavares I, Oliveira ME, Maia N, et al. Haplotype analysis of newly diagnosed Portuguese and Brazilian families with fibrinogen amyloidosis caused by the *FGA* p.Glu545Val variant. *Amyloid*. 2019;26(sup1):144-145. doi:10.1080/13506129.2019.1582500
3. Browning SR, Browning BL. Rapid and accurate haplotype phasing and missing-data inference for whole-genome association studies by use of localized haplotype clustering. *Am J Hum Genet*. 2007;81(5):1084-1097. doi:10.1086/521987
4. Reeve JP, Rannala B. DMLE+: Bayesian linkage disequilibrium gene mapping. *Bioinformatics*. 2002;18(6):894-895. doi:10.1093/bioinformatics/18.6.894

### Table I. Affected Families Haplotypes

| | Presumptive ancestral haplotype: | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | 14 | 8 | 18.1 | 23 | 18 | 7.3 | 19.1 | 12 | 13 | 14 | 11.2 | 15.2 | 12 |
| F01 | * | * | * | * | * | * | * | * | * | * | * | * | * |
| F02 | 9 | * | * | * | * | * | * | * | * | * | * | * | * |
| F03 | * | * | * | * | * | * | * | * | * | * | * | * | * |
| F04 | * | * | * | * | * | * | * | * | * | * | * | * | * |
| F05 | * | * | * | * | * | * | * | * | * | * | * | * | * |
| F06 | * | * | * | * | * | * | * | * | * | 13 | 9.2 | 8.2 | * |
| F10 | * | * | * | * | * | * | * | * | * | * | * | * | * |
| F12 | 13 | * | * | * | * | * | * | * | * | 12 | 9.2 | * | 13 |
| F07 | * | * | * | * | * | * | 18.1 | 11 | 12 | * | 6.2 | 8.2 | 13 |
| F08 | * | * | * | * | * | * | 18.1 | 11 | 12 | * | 6.2 | 8.2 | 13 |
| F09 | * | 10 | * | * | * | * | 18.1 | 11 | 12 | * | 6.2 | 8.2 | 19 |
| F11 | 12 | 9 | * | 24 | * | * | * | * | 12 | 13 | 6.2 | * | 19 |
| STRs | D4S1189i | D4S0594i | D4S2999 | D4S3021 | FGA | D4S0209i | D4S2976 | D4S1225i | D4S0183i | D4S3016 | D4S0232i | D4S1585 | D4S1498 |

* = same as presumptive ancestral haplotype

## Concluding Remarks

No genetic trio data was available. So while extended sibships and multigenerational data was included, the inferred haplotypes are not certain. An effort should be made to obtain the missing data.

Our results point to a relatively old mutation (2500 years), which could explain, at least in part, the wide dissemination of FGA p.Glu545Val.

It would be interesting to extend this study to other populations, to see if there is evidence for a common ancestor/multiple origins, and to try to establish a pattern of disease dissemination.