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THE DAMAGE DEGREE OF ANCIENT DNA OF *SUS SCROFA FERUS* AND *SUS SCROFA DOMESTICUS* REVEALED BY MOLECULAR MARKERS: CYT B GENE

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A big interest in animal domestication has developed over the 20th century, and the advances in the biomolecular technology shed light on certain aspects which previously could only be hypothesised. Pigs are among the most investigated domestic animals due to their significant economical value and the origin of pig domestication goes back in the past for about 10.000 years, in the Neolithic Period. From the beginning of the 20th century different theories related to the origin of domestic pigs have been formulated and they all relied mostly on the morphologycal data obtained from bone remains analysis. Only at the end of the century the biomolecular genetics approach starts to solve many of the misteries of pig domestication. The studies have concentrated so far on the analysis of mitochondiral DNA and, from this, only one molecular marker has been widely used: the D-loop fragment.

The present study aims to investigate the compatibility of another molecular marker, the cyt b gene, for the ancient DNA sequences required in the phylogenetic analysis of pig domestication.

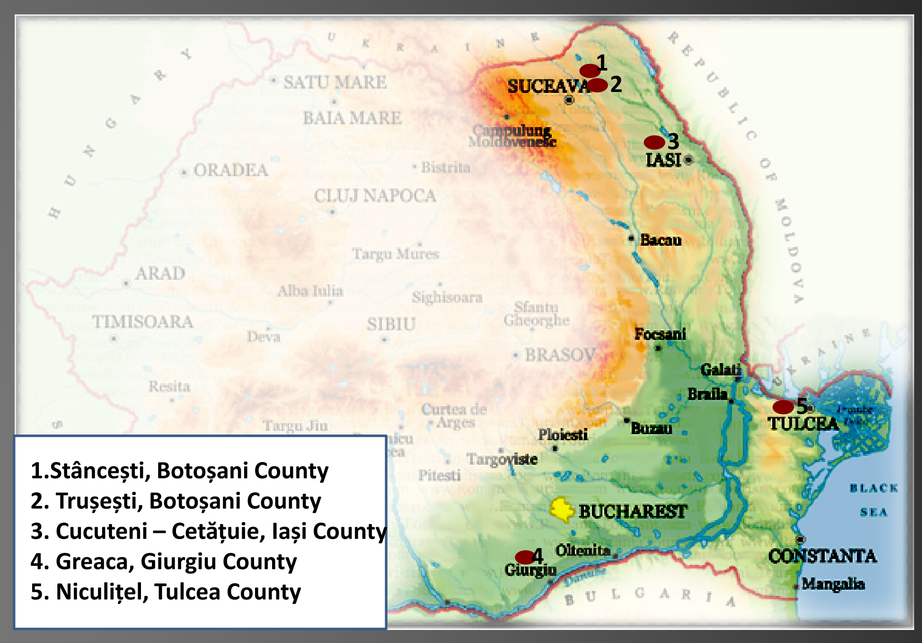
Material and methods

The 29 samples, representing bone remains, were collected from five different archaeologycal sites from the East and Southeast of Romania. The tissue was powdered and DNA was isolated and purified by the phenol: chloroform: isoamil acid protocol.

The origin, age and number of samples

Archaeologyc site	Samples age	Number of analysed samples
1. Trușești	Cucuteni Culture (4500 – 3500 BC)	5
2. Cucuteni-Cetățuie	Cucuteni Culture (4500 – 3500 BC)	5
3. Stăncești	VII-III century BC	15
4. Greaca	IV – I century BC	2
5. Niculițel	II-III century AD	2

Samples *of Sus scrofa domesticus* and *Sus scrofa ferus*



The distribution of archaeologyc sites on Romania's territory

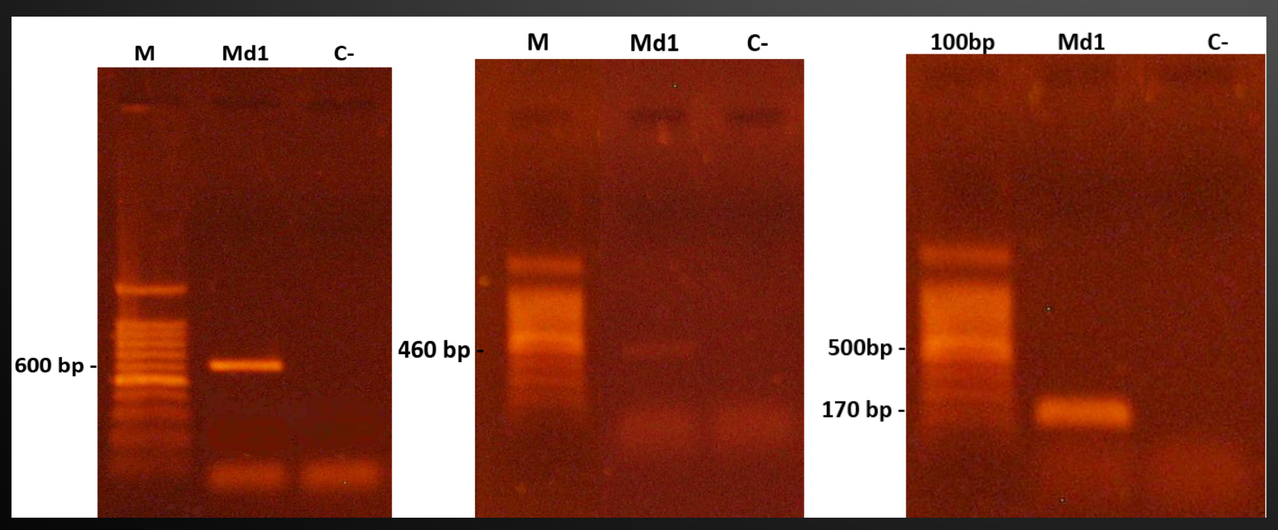
The aim of the study was to get a maximum length of the fragments of cyt b gene for as many samples as possible. Thus, three sets of primers were used for the PCR, starting with those for 600 bp, then for 460 bp, and in the end for 170 bp fragments. PCR was performed in a 25 µl reaction volume containing Go Taq Green Master Mix (Promega), direct and reverse primers, DNA and nuclease free water to 25 µl.

Results

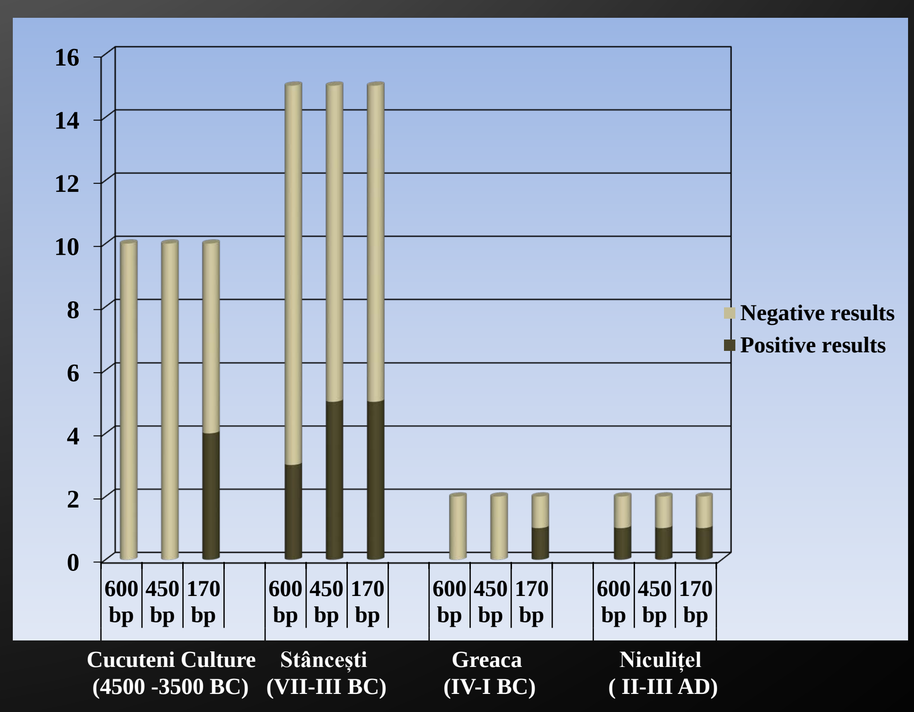
In the first stage, all samples were amplified with primers targeting a 600 bp DNAfragment . Only four DNA fragments from the 29 samples were obtained, but none of them was from the Cucuteni Culture period: three from the 15 samples from Stăncești (VII-III BC) and one of the two samples from Niculițel (II-III AD).

Secondly, the shorter DNA fragment, of 460 bp in length, was amplified for two more samples from Stâncești (VII-III BC).

Finally, a 170 bp DNA fragment was amplified in a total of 11 samples: 4 from the Cucuteni Culture, 5 from Stăncești, one from Greaca and one from Niculițel.



Comparative gel electrophoresis for one sample (from Niculițel) successfully amplified with the three pairs of primers



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