

POSTER: Dyes and proteins analysis in a unique workflow: a new methodology for archaeological textiles

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Archaeological textiles represent precious remains from ancient culture; this fact lies in the historical and cultural importance of the information that can be obtained by such relics. However, their extremely complicated state of preservation requires highly sensitive analytical tools to perform a comprehensive study. Starting from these considerations, this study focuses on both components of ancient wool textiles, dyes - especially anthraquinone ones- and keratins (IFs) and keratins associated proteins (KAPs). The Marie Skłodowska-Curie Actions PARCA project aims to develop an innovative protocol that would combine dye and protein analysis in a single extraction, minimizing the sampling while maximizing the amount of information obtained. Proteins and dyes from madder-dyed wool were extracted simultaneously using the novel TCEP/CAA method [1] or a modified urea method. After extraction, dyes were isolated with various clean-up methods, such as LLE, μ-SPE, or stage tips, evaluating their application in different steps of the protocol to minimize any possible alteration in the protein composition. Proteins were digested with trypsin either in-solution after dialysis or with SP3 [2] and desalted with C18 SPE/C18 stage tips. Peptides were separated with ThermoScientific Acclaim PepMap 100 trap and analytical columns at 300 nL/min; dyes with a Waters BEH Shield RP18 with an Ultimate 3000 at 200 uL/min and detected on an Orbitrap Elite.

The two protocols proved to be able to extract both components, even if with some differences. From the point of view of IFs and KAPs, the modified urea protocol was able to extract several type I and type II keratins together with several KAPs, with an average of 20 IFs protein groups and slightly fewer (15) for KAPs. The best match has a 75% protein coverage. Dyed and undyed samples were analyzed; the greater number of proteins obtained for undyed wool confirms that dyeing process impacts protein extraction and increases the deamidation values, a common marker of ageing process [3].

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This study opens also to new perspective for dyes analyses: the urea protocol was effective in extracting dyes in a sensitive shorter time than common extraction protocol for dyes in similar pH condition [4].

These protocols will be applied to charred samples from Pompeii, Vesuvian and Greek area [5].

The combined protein and dye extraction represents an important step in the analyses of archeological textiles, because it makes it possible to obtain a comprehensive analysis from a single unique sample.

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