

TRYPTIC PEPTIDE ANALYSIS OF PROTEIN BINDERS IN WORKS OF ART BY LIQUID CHROMATOGRAPHY – TANDEM MASS SPECTROMETRY

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Comprehension of the materials, such as binding media, used by artists is of uttermost importance in restoration and in art historical studies. The most frequently used binders are drying oils and proteins; in this study focus is placed on proteins. Most actual methods for protein binder identification are based on complete hydrolyzation of the protein matter into its amino acids and separation/detection with gas chromatography – mass spectrometry (GC-MS) or high performance liquid chromatography (HPLC) after derivatization. Because amino acids itself are not characteristic for a protein, identification is often based on the relative amount of 7 stable amino acids. In the current study a proteomics approach was used, in which the proteins were digested enzymatically into peptides using trypsin before being separated and detected by liquid chromatography – electrospray ionisation tandem mass spectrometry (LC-ESI-MS/MS). Mascot (Matrix Science) was used to analyze the resulting data and for protein identification. This way, amino acid sequences could be studied that retain much more information about the proteins, their degradation and pigment-binder interactions. The protein content of homemade paint samples was extracted using different methods and analysed to select the best extraction strategy based on the number of peptides that were identified. A large dataset of 4 binders (animal glue, egg yolk, egg white and casein), mixed with 10 common pigments with different chemical properties was used to study the influence of pigments on the extraction method. Analytical characteristics of the selected method were determined. Finally the method was applied to historic paint samples. The results were compared with those obtained by traditional amino acid analysis methods.