

MALDI-MS² ion trap quantification of Cu-GHK

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The project aims to develop novel, environmentally friendly, and cost-effective methods to evaluate the physical stability and degradation of molecules key to skincare products and treatments, while combating the limitations of quantifying low mass analytes through Matrix Assisted Laser Desorption Ionization mass spectrometry (MALDI). Using MALDI-Quadrupole Digital Ion Trap (MALDI-DIT), we will quantify in commercial products a molecule essential to skin tissue health: copper tripeptide-1. Copper Tripeptide-1, otherwise known as Cu-GHK (sequence glycine-histidine-lysine) is a biomimetic peptide resembling the alpha chain of collagen. Cu-GHK has various functions regarding human skin; the peptide GHK chelates with Cu^{2+} , acting as a carrier peptide to transport Cu^{2+} ions to skin epithelial cells. Additionally, Cu-GHK transports Cu^{2+} ions to lysyl oxidase. ¹Lysyl oxidase oxidizes lysyl residues in collagen and elastin; the proteins found in the extracellular matrix and are responsible for maintaining connective tissue health and the elasticity of skin. Degradation of elastin has been linked to skin aging, and so quantification methods for Cu-GHK are highly valuable.¹

Due to Cu-GHK's ability to transport Cu^{2+} ions to lysyl oxidase, Cu-GHK is linked to maintaining skin integrity and antiaging and is commonly used in commercial skincare. However, there are no established studies of the absolute quantification of Cu-GHK proper, let alone in cosmetic formulations using mass spectrometry. Likewise quality testing is lacking, as in In the U.S, cosmetic products are regulated by the FDA, but this is limited to prohibiting cosmetics that are adulterated with poisonous, putrid, or decomposed substances, and prohibiting false and misleading labeling.² Cosmetics, which include topical creams and serums that have Cu-GHK, do not require FDA pre-market approval, and it is on the companies and individuals to verify their products contain the listed ingredients.² Due to the lack of quality testing and quantification, there is a great need for rigorous, independent analytical validation of Cu-GHK in cosmetics.

We intend to first develop and validate a quantitative method using MALDI-QIT for Cu-GHK using the inexpensive analogue Cu-AHK as internal standard. MALDI-QIT is cost effective and low-waste, requiring ≤ 1 uL of target sample and 2-20 μL of matrix, in direct contrast to LC-MS which requires large amounts of toxic organic solvents. There are few preparation steps for MALDI compared to other methods such as LC-MS, and it typically produces less fragmented mass spectra compared to ionization methods such as ESI.³ MALDI-QIT will be used to assess the linearity, limit of quantification (LOQ), limit of detection (LOD) of Cu-GHK, as well as recovery studies and matrix effect of commercial products. We will then use these established parameters to quantify Cu-GHK in commercial skincare products.

We have generated three precise and reproducible calibration curves and achieved a low limit of detection and quantification of Cu-GHK. Through interpolation, we found that Cu-GHK has been reliability quantified in the commercial product Asterwood in three reproducibility trials and is recovered well in both Asterwood and blank serums. Additionally, the matrix effect of Asterwood was shown to be negligible. In the future, we aim to quantify Cu-GHK in other serums, and validate MALDI quantification accuracy with LC-MS.

References

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