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Validation of an UHPLC method for the determination of hydroxymethylfurfural in honey

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Background/PURPOSE: The increasing global demand for honey has intensified concerns about its quality and authenticity. One of the primary indicators of honey adulteration and deterioration is hydroxymethylfurfural (HMF), a compound formed during excessive heating or prolonged storage. In Panama, the lack of validated analytical methods for HMF quantification has limited the ability to assess honey quality reliably. This study aimed to validate an Ultra High-Performance Liquid Chromatography (UHPLC) method for the accurate determination of HMF in commercial honey.

Problem Description: Honey adulteration compromises consumer health and market integrity, affecting both local beekeepers and regulatory agencies. The absence of a validated method in Panama has made it difficult to monitor HMF levels effectively, thus hindering quality control efforts.

METHODOLOGY: A UHPLC method with diode-array detection (UHPLC-DAD) was validated following the International Council for Harmonisation (ICH) guidelines. The method employed solid-phase extraction (SPE) for sample cleanup, followed by chromatographic analysis using a C18 column and a water-methanol mobile phase. Linearity, specificity, precision, accuracy, limit of detection (LOD), and limit of quantification (LOQ) were assessed to ensure method reliability.

Results and CONCLUSION: The method demonstrated excellent specificity, with no matrix interferences detected. It showed robust linearity ($R^2 = 0.9995$) within the analytical range of 0.50–15.23 µg/mL. The precision tests confirmed repeatability and intermediate precision within acceptable limits. Accuracy was verified through recovery assays (96.7%–101.7%), and the method achieved an LOD of 0.15 µg/mL and an LOQ of 0.45 µg/mL. The validated UHPLC-DAD method was successfully applied to analyze commercial and artisanal honey samples, confirming its suitability for routine quality control and regulatory applications.

Practical Implications for Beekeepers: This validated method provides beekeepers and regulatory agencies with a robust analytical tool to assess honey quality, ensuring compliance with international standards. By identifying adulteration and improper storage conditions, beekeepers can enhance product quality, maintain consumer trust, and access broader markets with higher confidence.