

Introduction

Coffee, a widely consumed beverage, is valued for both its flavor and bioactive compounds. Among these, quercetin and its derivatives—classified as flavonoids—are known for their potential health benefits, including antioxidant, anti-inflammatory and neuroprotective properties ^{1,2}. While quercetin is found in green coffee beans ³, its presence in roasted coffee and popular beverages like espresso and moka is less understood. This study focuses on quantifying six quercetin derivatives in *Coffea arabica* beans from five geographical origins (Brazil, Colombia, Ethiopia, Guatemala, India), prepared using two brewing methods (espresso, moka). Additionally, the impact of different post-harvest processing methods (Natural, Washed, Honey) on the quercetin content is examined.

Materials and Methods

Coffee samples:
Roasted *Coffea arabica* samples from five origins: *Brazil, Colombia, Ethiopia, Guatemala and India* were provided by Illycaffè S.p.A. Three samples from *Guatemala* (Natural, Washed, Honey) were provided by Perfero Caffè.

Extraction procedures:
Three methods were evaluated to optimize the recovery of quercetin and its derivatives from roasted coffee:

Acidic hydrolysis: 1 g of roasted coffee was treated with 10 mL of 70% ethanol at pH 2 (adjusted with 2N HCl), sonicated at 60°C for 90 minutes, followed by centrifugation at 5000 rpm for 10 minutes.

Alkaline hydrolysis: similar to acidic hydrolysis but with pH 9 (adjusted with NaOH).

Direct solvent extraction: 70% ethanol with no pH adjustment, sonicated at 25°C for 90 minutes.

Coffee beverage preparation:
Espresso (EC): prepared using a VA388 Black Eagle Maverick machine, 25 ± 2 mL of espresso was extracted from 9 g of ground coffee at 93°C.

Moka (MC): 250 mL of water and 25 g of ground coffee were used in a MOKA EXPRESS® coffee maker. The preparation lasted about 10-15 minutes.

Quantification of quercetin derivatives:
Analyses were determined using HPLC-ESI-MS/MS, equipped with a Synergy polar-RP 80Å analytical column. Separation was achieved using a gradient elution with water and methanol (both with 0.1% formic acid) ³.

Six quercetin derivatives were quantified: quercetin, quercetin-3-glucuronide, rutin, hyperoside, quercitrin and isorhamnetin.

Statistical analysis:
All experiments were performed in triplicate and differences in analyte concentrations were evaluated using one-way ANOVA with post-hoc Tukey HSD tests (*p* < 0.05).

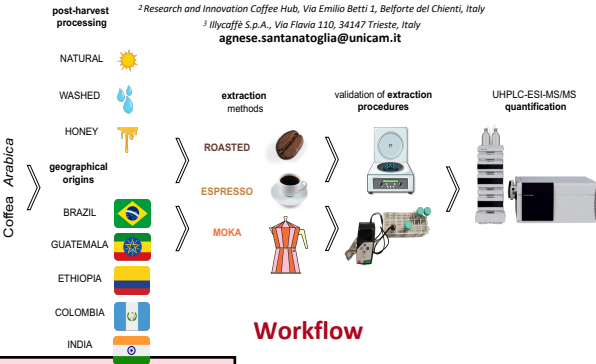
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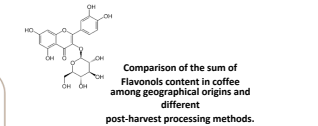
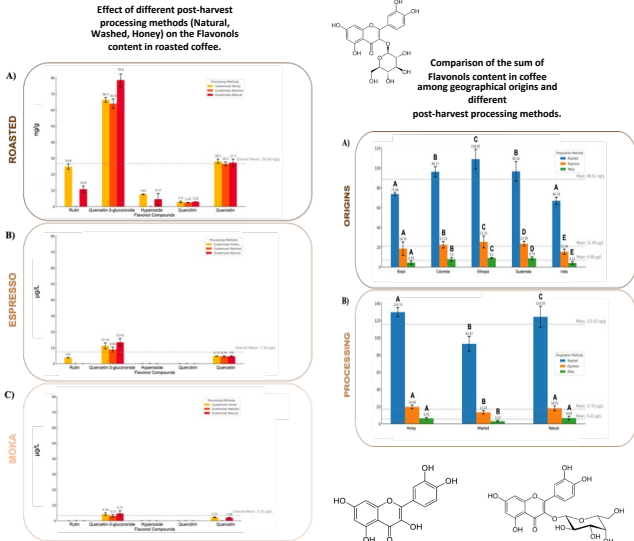
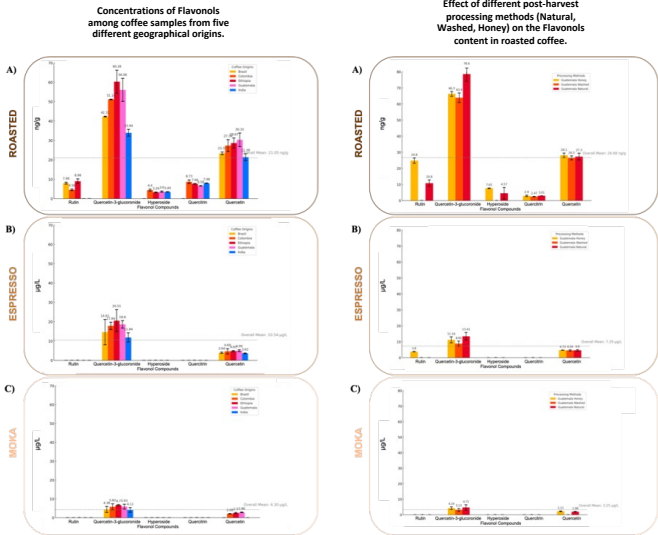
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Workflow



Results

Ethiopian coffee had the highest total quercetin derivatives content, with **108.85 ± 10.03 ng/g**, primarily quercetin-3-glucuronide (**60.28 ± 5.95 ng/g**). **Guatemalan and Colombian** coffees also exhibited high levels of quercetin derivatives, with total contents of **96.56 ± 9.89 ng/g** and **96.17 ± 5.02 ng/g**, respectively. **Indian and Brazilian** coffees had comparatively lower levels of flavonoids, particularly quercetin-3-glucuronide. **Honey** processed coffee from Guatemala showed the highest total quercetin content (**129.75 ± 5.1 ng/g**), followed by **Natural** processing (**124.28 ± 12.2 ng/g**). **Washed** coffee had the lowest quercetin content across all geographical origins, suggesting that *less invasive* processing methods (e.g., Natural and Honey) preserve quercetin derivatives more effectively. **Espresso** extraction yielded **higher flavonoid content** than **Moka**, particularly for Ethiopian (**25.32 ± 5.95 µg/L**) and Guatemalan (**23.59 ± 2.35 µg/L**) coffees ⁴. Total flavanol content was consistently higher in espresso samples across all origins compared to moka, likely due to the higher pressure and temperature used in espresso preparation.