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Maceration vs. Ultrasound extraction of phenolic compounds from *Bridelia ferruginea* Benth. (Phyllanthaceae): effects of solid-liquid ratio

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Abstract
Phenolic plant metabolites are bioactive compounds with significant antioxidant and therapeutic potential, making them valuable for pharmaceutical and nutraceutical applications. Bridelia ferruginea, a medicinal plant widely used in traditional medicine, is a promising source of these compounds. However, their extraction efficiency depends critically on the methodology and solvent-to-solid ratio employed. This study systematically compared conventional maceration (MAC) and ultrasound-assisted extraction (UAE) for recovering polyphenols and flavonoids from B. ferruginea stem bark, while evaluating the influence of solvent-to-solid ratios (1/60, 1/80, 1/120 g/mL). Total phenolic compounds and flavonoids contents were quantified spectrophotometrically using the Folin-Ciocalteu and aluminum chloride ($AlCl_3$) assays, respectively. Statistical analysis (ANOVA with Tukey's post-hoc test, $p < 0.05$) revealed that UAE significantly enhanced extraction yields compared to maceration. The optimal phenolic yield (291.5 ± 6.3 mg GAE/g DM) was achieved at a 1/80 ratio using UAE, whereas the highest flavonoid content (17.6 ± 1.1 mg QE/g DM) was obtained at 1/60 g/mL. Excessive solvent volume (1/120 g/mL) led to reduced yields, likely due to dilution effects. These findings demonstrate that UAE is a superior technique for extracting bioactive compounds from B. ferruginea, with extraction efficiency strongly influenced by the solvent-to-solid ratio. This study provides actionable insights for optimizing the recovery of phytochemicals from medicinal plants, supporting their potential application in health-focused industries.

Keywords*: Bridelia ferruginea, ultrasonic-assisted extraction, maceration, phenolic compounds, solid-liquid ratio.*

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I. Introduction

Natural products and their bioactive compounds, particularly phytophenols (plant phenolics), are attracting growing interest due to their antioxidant, anti-inflammatory, and antimicrobial properties [1,2]. Among promising plant species, *Bridelia ferruginea* stands out for its traditional uses in treating intestinal disorders, urinary infections, dental caries, and oral ailments [3]. These empirical applications, supported by recent phytochemical studies [4,5], highlight its therapeutic potential and justify further exploration of its active compounds.

The efficiency of extracting these molecules heavily depends on the method used. While maceration remains a conventional technique [6], innovative approaches such as ultrasound-assisted extraction (UAE) offer notable advantages: reduced processing time, improved yields, and solvent economy [7,8]. However, optimizing operational parameters—particularly the solid-to-liquid ratio—is crucial to maximize the extraction of polyphenols and flavonoids.

In this study, we systematically compare conventional maceration and UAE for extracting phenolic compounds from *B. ferruginea* bark. Three solid-to-liquid ratios (1/60, 1/80, and 1/120) are evaluated under standardized conditions (60% acetone, 40°C, 40 min) to identify optimal extraction conditions. The extracts are quantitatively analyzed, and data are processed using one-way ANOVA followed by Tukey's post-hoc tests to ensure statistical robustness.

This methodological approach aims not only to assess the comparative efficiency of the two techniques but also to provide a scientific basis for optimizing extraction processes, with potential implications for pharmaceutical and nutraceutical applications.

II. Material And Methods

Plant material and chemicals

Fresh bark samples from the trunks of *Bridelia ferruginea* were collected in the Kani region (northern Côte d'Ivoire, geographical coordinates: 8°28'59'' N, 6°35'58'' W) in accordance with ethical and botanical standards. Botanical identification was conducted by an expert at the National Floristic Center of Côte d'Ivoire. The samples were air-dried at room temperature and then ground into a fine powder using a mortar. Analyses were conducted using high-purity reagents, including Folin-Ciocalteu reagent (Sigma-Aldrich, St. Louis, USA) for total phenol quantification, as well as aluminum chloride ($AlCl_3$) and sodium carbonate (Na_2CO_3) for phytochemical analyses. Analytical-grade solvents, including methanol, ethanol, and ultrapure water, were used for the various treatments. All other standard reagents were also sourced from Sigma-Aldrich.

Extraction Procedures

General Conditions: all extractions were carried out at 40 °C for 40 minutes, using three different solid/liquid ratios (1/60, 1/80, and 1/120, w/v) to evaluate their influence on yield. Each experiment was performed in triplicate to ensure the reproducibility of results.

Conventional Maceration Extraction: a precise quantity of 5.0 g of *Bridelia ferruginea* bark powder was placed in a conical flask and mixed with 60% acetone according to the tested solid/liquid ratios (300 mL for 1:60, 400 mL for 1:80, and 600 mL for 1/120). The sealed flask was placed in a thermostatic water bath at 40 °C with intermittent agitation (every 10 minutes) to optimize solvent diffusion. After 40 min, the mixture was filtered through Whatman No. 1 paper, and the filtrate was collected for analysis.

Ultrasound-Assisted Extraction: the same quantity of bark powder (5.0 g) was subjected to ultrasound-assisted extraction (40 kHz, 240 W) in 60% acetone, maintaining a constant temperature of 40 °C for 40 minutes. Ultrasonic waves induced cavitation, improving cell wall rupture and compound release. To prevent thermal degradation, the temperature was continuously monitored. After extraction, the mixture was decanted, filtered (Whatman No. 1), and the filtrate was stored for analysis.

Quantification of phenolic compounds and flavonoids

Determination of total phenols (Folin-Ciocalteu method): the total phenol content (TPC) was determined following the protocol of [9]. One milliliter of extract was mixed with 500 µL of Folin-Ciocalteu reagent (diluted to 10%). After 5 min, 1 mL of saturated Na_2CO_3 solution was added, followed by incubation for 30 min in the dark. Absorbance was measured at 765 nm using a UV-Vis spectrophotometer. A calibration curve (gallic acid, 0–100 mg/L) was used to express the results in mg GAE/g DM (gallic acid equivalents per gram of dry matter).

Determination of total flavonoids ($AlCl_3$ method): total flavonoids (TF) were quantified using the aluminum chloride method described by [10]. Two milliliters of extract were mixed with 1 mL of 5% $AlCl_3$ solution, incubated for 30 min at room temperature, and measured at 434 nm. Quercetin (0–50 mg/L) was used as the standard, and the results were expressed in mg QE/g DM (quercetin equivalents per gram of dry matter).

Data analysis and statistical methods

All analyses were performed in triplicate, with results expressed as mean ± standard deviation (SD). Differences in extraction yields (polyphenols and flavonoids) based on solid/liquid ratios (1/60, 1/80, 1/120 g/mL) and extraction methods (maceration vs. ultrasound) were evaluated using one-way ANOVA. When ANOVA indicated significant differences ($p < 0.05$), Tukey's post-hoc test was applied for pairwise comparisons. Statistical analyses and graph generation were carried out using RStudio (version 2024.12.1 Build 563). Results are presented as bar charts with error bars (representing SD) and summary tables of extraction yields for each experimental condition.

III. Results and Discussion

Analysis of Extraction Conditions

The analysis of extraction conditions (**Table no 1** and **Figures no 1-3**) reveals that ultrasound-assisted extraction (UAE) yields significantly higher total phenols (107.2–291.5 mg GAE/g DW) and flavonoid (6.2–17.6 mg QE/g

DM) contents compared to maceration (81.4–256.6 mg GAE/g DM and 3.8–12.8 mg QE/g DM, respectively), regardless of the solid-liquid ratio. This enhancement in yields is attributed to the ultrasonic cavitation effect, which promotes cell wall disruption and the release of bioactive compounds, as reported in previous studies [11–13]. These findings confirm the potential of UAE as an optimal extraction method for these secondary metabolites.

Table no 1: Summary of extraction conditions and total phenolic and flavonoid contents

Method	Solid-Liquid Ratio (g/mL)	Phenolic content (mg GAE/g DM)	Flavonoid content (mg QE/g DM)
Maceration	1/60	246.9 ± 4.8	12.8 ± 0.9
Maceration	1/80	256.6 ± 6.6	4.4 ± 0.3
Maceration	1/120	81.4 ± 3.2	3.8 ± 0.2
Ultrasound	1/60	285.7 ± 5.5	17.6 ± 1.1
Ultrasound	1/80	291.5 ± 6.3	6.2 ± 0.5
Ultrasound	1/120	107.2 ± 4.3	5.3 ± 0.3

The results demonstrate that both the extraction method and solid-liquid ratio significantly influence phenolic and flavonoid contents. Ultrasound-assisted extraction (UAE) proved more efficient than maceration, yielding maximum phenolic content (291.5 ± 6.3 mg GAE/g DM) at a 1/80 g/mL ratio, compared to 256.6 ± 6.6 mg GAE/g DM for maceration under the same conditions ($p < 0.05$). This improvement is attributed to ultrasound's cavitation effect, which disrupts cell walls and enhances phenolic compound release[14,15]. In contrast, flavonoid content reached its peak (17.6 ± 1.1 mg QE/g DM) with UAE at 1/60, suggesting these compounds require less solvent for optimal extraction. Analysis of the solid-liquid ratio reveals that 1/80 represents the optimal compromise for phenolic extraction, balancing plant material quantity with solvent volume. A lower ratio (1/60) limits compound diffusion due to insufficient solvent volume, while a higher ratio (1/120) causes excessive dilution, reducing extraction efficiency. These findings confirm the importance of optimizing extraction parameters to maximize bioactive compound yields. These findings align with previous research by [16], which established that moderate solid-liquid ratios optimize polyphenol extraction from *Camellia sinensis* when using eco-friendly extraction methods, while excessive solvent volumes lead to reduced compound concentrations. Complementary evidence comes from [17], whose work on mate tea leaves confirmed that UAE significantly improves polyphenol recovery, with peak yields consistently achieved at intermediate solvent volumes rather than extreme ratios.

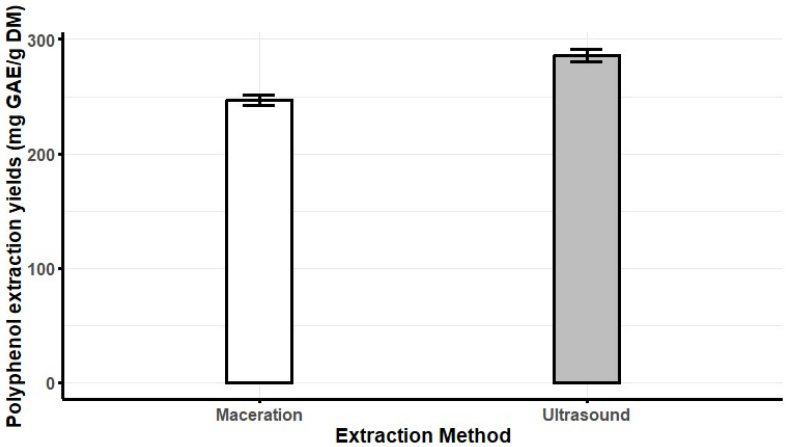

Figure no 1: Phenol yield (mg GAE/g DM) according to the extraction method – S/L Ratio 1/60

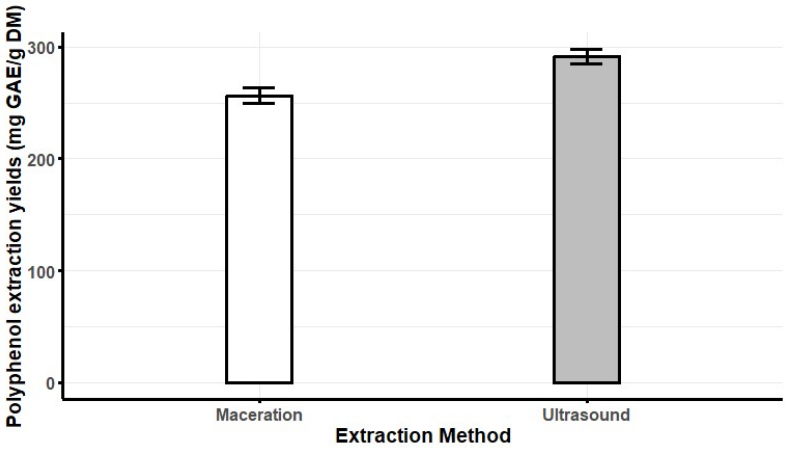

Figure no 2 : Phenol yield (mg GAE/g DM) according to the extraction method – S/L Ratio 1/80

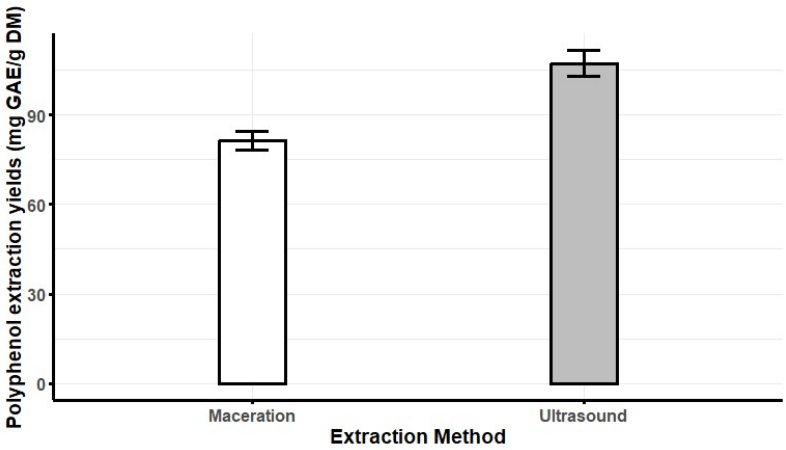

Figure no 3 : Phenol yield (mg GAE/g DM) according to the extraction method – S/L Ratio 1/120

Moreover, the results reveal a distinct trend for flavonoids (**Figures 4-6**), with maximum yield **(**17.6 ± 1.1 mg QE/g DM) achieved through UAE at a 1/60 ratio, followed by a progressive decline at higher ratios (1/80 and 1/120). This decrease suggests flavonoids exhibit greater sensitivity to dilution effects, where excess solvent reduces solid-solvent interactions, thereby limiting extraction efficiency. Unlike polyphenols, flavonoids appear to require more concentrated conditions for optimal recovery. These findings align with observations by [18] on *Ginkgo biloba*, where low solid-liquid ratios (1/20–1/50) enhanced flavonoid extraction, while excessive solvent volumes led to compound degradation or inefficient solubilization. The superiority of UAE over maceration stems from its mechanical action (cavitation), which disrupts cell walls more effectively than passive diffusion. This is supported by [19] in *Lactuca indica L.*, where ultrasound delivered significantly higher flavonoid yields than conventional methods.

Figure no 4 : Flavonoid yield (mg QE/g DM) according to the extraction method – S/L Ratio 1/60

Figure no 5 : Flavonoid yield (mg QE/g DM) according to the extraction method – S/L Ratio 1/80

Figure no 6 : Flavonoid yield (mg QE/g DM) according to the extraction method – S/L ratio 1/120

Comparison and statistical interpretation

Statistical analysis (ANOVA) revealed that the extraction method and solid-to-liquid ratio significantly influenced the yields of polyphenols and flavonoids ($p < 0.05$) (**Figures no 7-8**). Post-hoc comparisons indicated that ultrasound-assisted extraction (UAE) with a ratio of 1:80 yielded significantly higher polyphenol levels, while a ratio of 1/60 was optimal for flavonoids. These results highlight the advantages of UAE over maceration, particularly in terms of reduced extraction time and improved compound recovery. However, they also reveal differences in the behavior of polyphenols and flavonoids based on the volume of solvent used. While polyphenols achieve maximum yield with a moderate solid-to-liquid ratio, promoting better diffusion, flavonoids exhibit more efficient extraction at lower ratios, likely due to their solubility properties and sensitivity to dilution. These observations align with existing literature and emphasize the need to optimize extraction conditions based on the targeted compounds. Future research could explore the influence of other parameters, such as temperature and solvent nature, to further enhance the efficiency of extraction processes.

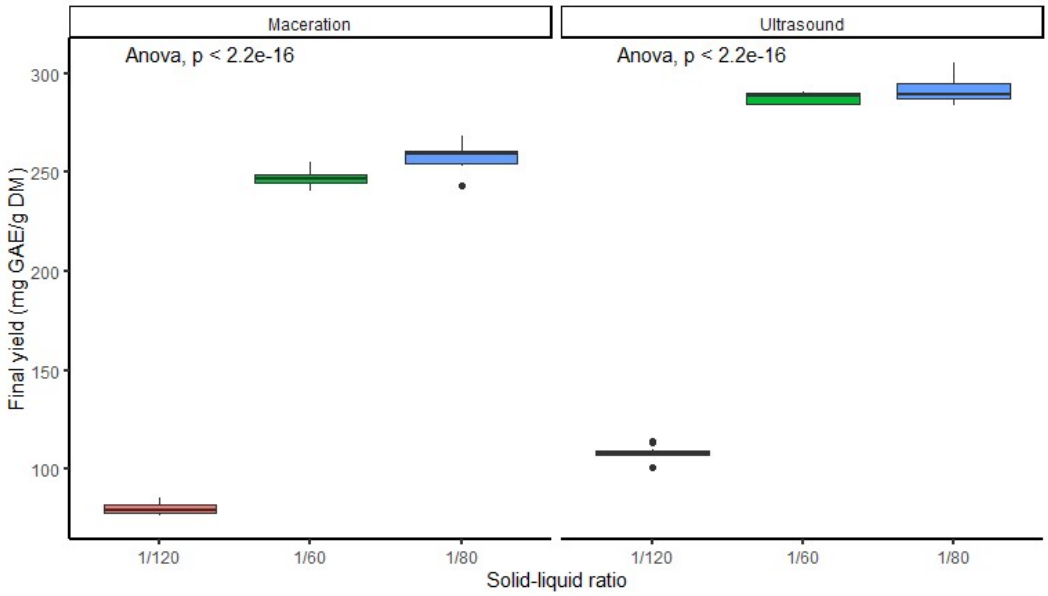

Figure no 7 : Statistical comparison of phenolic compound extraction yields between maceration and ultrasound for each solid-liquid ratio

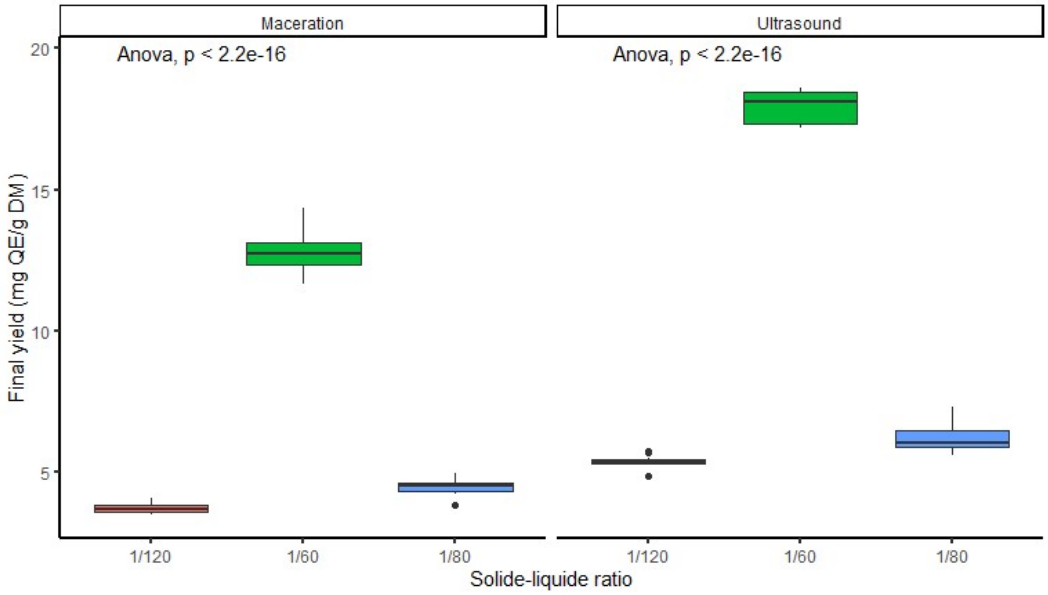

Figure no 8 : Statistical comparison of flavonoid extraction yields between maceration and ultrasound for each solid-liquid ratio

IV. Conclusion

This study revealed the influence of the extraction method and solid-to-liquid ratio on the extraction of phenolic compounds and flavonoids from the bark of *Bridelia ferruginea*. The results showed that ultrasound-assisted extraction (UAE) was significantly more effective than maceration, confirming its potential to enhance total phenol and flavonoid yields. Maximum yield of total phenols (291.5 ± 6.3 mg GAE/g DM) was achieved with UAE using a solid-to-liquid ratio of 1:80, while the highest flavonoid yield (17.6 ± 1.1 mg QE/g DM) was obtained with a ratio of 1/60. These findings indicate that phenolic compounds and flavonoids respond differently to extraction parameters, highlighting the importance of tailoring the process based on the properties of each compound. UAE has proven to be a promising technique to replace traditional maceration, reducing extraction time while increasing the recovery of bioactive molecules. Further studies could investigate additional factors, such as temperature, solvent nature, and extraction kinetics, to further optimize the process. Moreover, validating these results on a larger scale and with various plant matrices would strengthen the industrial applicability of UAE for extracting bioactive compounds from medicinal plants.

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