JPPIPA 11(8) (2025)

Jurnal Penelitian Pendidikan IPA

http://jppipa.unram.ac.id/index.php/jppipa/index

Optimization of Ultrasonic-Assisted Extraction of Curcuminoids from *Temulawak* (Curcuma xanthorrhiza Roxb.) Using Response Surface Methodology

Yusnira[1*], Kasman Ediputra[2]

[1] Department of Nutritions, Faculty of Health Science, Universitas Pahlawan Tuanku Tambusai, Riau, Indonesia.
[2] Department of Mathematics Education, Faculty of Education, Universitas Pahlawan Tuanku Tambusai, Riau, Indonesia.

Received: May 16, 2025
Revised: July 10, 2025
Accepted: August 25, 2025
Published: August 31, 2025

Corresponding Author:
Yusnira
yusnira73@gmail.com

DOI: 10.29303/jppipa.v11i8.12108

Abstract: *Temulawak* (Curcuma xanthorrhiza Roxb.) is a native Indonesian medicinal plant that has a lot of curcuminoids with strong pharmacological activity. However, traditional extraction procedures don't always work well or provide good quality extracts. The goal of this research is to find the best way to get curcuminoids out of *temulawak* utilizing ultrasonic-assisted extraction (UAE) and response surface methodology (RSM). The Box-Behnken Design was used to optimize the process with three independent variables: ultrasonic power (200–400 W), extraction duration (20–60 minutes), and the ratio of material to solvent (1:15–1:35 g/mL). We used validated HPLC to look at curcuminoids, the DPPH technique to measure antioxidant activity, and a second-order polynomial model to look at the data. The mathematical model shows a good fit (R^2 = 0.9847), with all variables having a significant impact on the curcuminoid yield. The best results were achieved with an ultrasonic power of 354 W, an extraction time of 52 minutes, and a material to solvent ratio of 1:22 g/mL, leading to a total curcuminoid yield of 23.4±0.6 mg/g of simplicia and an antioxidant activity IC_{50} of 31.2±1.1 µg/mL. The enhanced UAE process gave 8.4 times more yield than maceration and took 28 times less time to extract. The improved UAE is a quick and long-lasting way to get curcuminoids out of *temulawak* with a high yield and high-quality extract. This study gives scientists the information they need to create turmeric extraction technologies on an industrial scale to help Indonesia's pharmaceutical and nutraceutical businesses.

Keywords: Curcuminoids; Response surface methodology; *Temulawak*; Ultrasonic-assisted extraction.

Introduction

Temulawak (Curcuma xanthorrhiza Roxb.) is one of Indonesia's native medicinal plants belonging to the Zingiberaceae family and has been utilized for generations as a raw material for traditional herbal medicine and traditional medicine (Paryanto & Srijanto, 2006; Rahmat et al., 2021). The plant known by the local names koneng gede (Sunda), temu labak (Madura), and tommo (Bali) thrives in the tropical regions of Indonesia, particularly on the islands of Java, Sumatra, Kalimantan, and Maluku, and has spread to several Southeast Asian countries such as Malaysia, Thailand, and the Philippines (Rosidi et al., 2014; Simamora et al., 2024).

Traditionally, *temulawak* has been used to address various health issues such as loss of appetite, digestive disorders, liver diseases, constipation, bloody diarrhea, dysentery, arthritis, fever in children, hypotriglyceridemia, hemorrhoids, leucorrhea, rheumatism, and skin disorders (Rahmat et al., 2021;

How to Cite:
Yusnira, & Ediputra, K. (2025). Optimization of Ultrasonic-Assisted Extraction of Curcuminoids from Temulawak (Curcuma xanthorrhiza Roxb.) Using Response Surface Methodology. *Jurnal Penelitian Pendidikan IPA*, *11*(8), 183–192. https://doi.org/10.29303/jppipa.v11i8.12108

Setyowati & Suryani, 2013). The healing benefits of *temulawak* have been proven by several studies that show it has effects like reducing inflammation, fighting bacteria, acting as an antioxidant, protecting the brain and kidneys, fighting tumors, and protecting the liver, and it has been sent to Europe for use since at least 1963, especially for treating upset stomach, infections, and skin and liver problems.

The main bioactive compounds contained in the rhizome of *temulawak* are curcuminoids, which consist of curcumin, demethoxycurcumin, and bisdemethoxycurcumin as components responsible for the characteristic yellow color and pharmacological activity (Binello et al., 2020; Mujahid et al., 2011). Curcuminoids are quality parameters established in the Indonesian Herbal Pharmacopoeia for *temulawak* simplicia because these compounds have scientifically proven antioxidant, anti-inflammatory, antimicrobial, and anticancer activities (Shirsath et al., 2017). Besides curcuminoids, *temulawak* also has a special compound called xanthorrhizol, which is a type of sesquiterpene not found in other Curcuma plants and adds unique health benefits, including anticancer, antimicrobial, anti-inflammatory, antioxidant, antihyperglycemic, antihypertensive, neuroprotective, and estrogenic effects.

The curcuminoid content in the rhizome of *temulawak* ranges from 1-3%, with the composition of curcumin as the main component (75%), desmethoxycurcumin (10-20%), and bisdesmethoxycurcumin (5%) of the total curcuminoids. These active components have strong antioxidant activity, with a Radical Scavenging Activity (%RSA) percentage reaching 80.11% in *temulawak* extract extracted with a powder:ethanol ratio of 1:5 (Patil et al., 2021; Setyowati & Suryani, 2013). Conventional extraction methods such as maceration, reflux, and Soxhlet, which have been used to extract curcuminoids from *temulawak*, have several significant drawbacks that hinder the efficiency of the process (Kiamahalleh et al., 2016; Mujahid et al., 2011). The reflux method recommended by the Indonesian Herbal Pharmacopoeia for determining curcuminoid content requires special equipment, a relatively long time (8-12 hours), and a considerable amount of energy and chemicals, making it less practical and efficient (Binello et al., 2020; Mujahid et al., 2011). Additionally, traditional methods use high temperatures that can break down curcuminoids, require a lot of organic solvents, and take a long time to extract, leading to low efficiency and high energy use. Research by (ahyono et al. (2011) found that drying *temulawak* rhizomes at high temperatures can change the amount and makeup of curcuminoids, and another study showed that the Soxhlet method only extracted 62% of curcuminoids in 8 hours, while newer methods can get 72% in just 1 hour (Patil et al., 2021; Shirsath et al., 2017).

The new extraction technology has created the Ultrasonic-Assisted Extraction (UAE) method, which is a more effective and eco-friendly way to extract useful compounds from natural materials. UAE uses ultrasonic waves that range from 20 kHz to 2000 kHz to create tiny bubbles that form, grow, and burst, which damages plant cell walls and helps release the compounds we want to extract. The UAE mechanism involves shear forces generated from cavitation along with shock waves that induce physical damage to the plant cell walls, thereby facilitating the release of the compounds to be extracted, where solvent diffusion is also enhanced by the swelling of the plant matrix and hydration by the solvent (Patil et al., 2021; Rusli et al., 2024). The advantages of UAE compared to conventional methods include shorter extraction times (from hours to minutes), reduced solvent usage, lower operating temperatures (30-60°C), and the ability to maintain the structural integrity of thermolabile compounds such as curcuminoids (Liao et al., 2015; Shirsath et al., 2017).

Research shows that UAE can achieve a maximum yield of 72% when the temperature is set to 35°C, the solid-to-solvent ratio is 1:25, the particle size is 0.09 mm, the ultrasonic power is 250 W, and the frequency is 22 kHz, using ethanol as the solvent. Response Surface Methodology (RSM) has proven to be a powerful statistical and mathematical tool for optimizing extraction processes because it can evaluate the effects of multiple factors and their interactions on one or more response variables (Aydar, 2018; Hartuti & Supardan, 2013). RSM uses mathematical and statistical techniques to model the relationship between independent variables (factors) and dependent variables (responses) through an empirical approach first introduced by Box and Wilson in 1951 (Olabinjo et al., 2020). This method allows researchers to identify optimal operating conditions, predict responses in the experimental domain, and offer explanations for factor interactions that may not be visible through the conventional one-factor-at-a-time approach (Chelladurai et al., 2021; Rusli et al., 2024). Central Composite Design (CCD) and Box-Behnken Design (BBD) are the most popular experimental methods used in RSM for optimizing extractions because they allow for effective exploration of the response surface while requiring fewer experiments. The application of RSM in the optimization of bioactive compound extraction has been successfully implemented on various plants, such as the optimization of ginger oleoresin extraction using ultrasonic waves, which successfully identified the optimum conditions at a ginger powder:ethanol ratio of

1:3.70, a temperature of 46°C, and a time of 129 minutes, resulting in a yield of 8.884% and a refractive index of 1.487 (Hartuti & Supardan, 2013), as well as the optimization of curcuminoid extraction from Curcuma longa using a deep eutectic solvent, achieving a maximum yield of 77.13 mg/g (Patil et al., 2021).

Although research on the extraction of curcuminoids from various Curcuma species has advanced rapidly, there remains a significant research gap in optimizing extraction parameters specifically for *temulawak* (Curcuma xanthorrhiza Roxb.) using advanced statistical methodologies (Bezerra et al., 2008; Kawiji et al., 2011; Patil et al., 2021). Previous research conducted by Simamora et al. (2024) focused more on the optimization of xanthorrhizol extraction using a natural deep eutectic solvent with the RSM-Box Behnken design, which resulted in a yield of 17.62 mg/g under optimal conditions of 30% water content in GluLA, an S/L ratio of 1/15 g/mL, and an extraction time of 20 minutes. In contrast, comprehensive optimization of curcuminoid extraction from *temulawak* using UAE with the RSM approach is still very limited. The complexity of the chemical composition of *temulawak*, with its unique combination of curcuminoids and other bioactive compounds, requires a systematic approach in extraction optimization that considers multiple process variables simultaneously (Kawiji et al., 2011; Rahmat et al., 2021). Additionally, the potential degradation of curcuminoids under harsh extraction conditions and the need for a sustainable extraction process emphasize the importance of developing optimized UAE protocols to maximize yield while maintaining compound integrity (Binello et al., 2020; Mujahid et al., 2011). Using RSM to improve UAE methods for extracting curcuminoids from *temulawak* is an important area of research that can help create a more efficient, cost-effective, and eco-friendly way to extract this valuable Indonesian medicinal plant.

Current research conditions indicate several critical challenges in the extraction of curcuminoids from *temulawak* that require systematic investigation. Traditional extraction methods suffer from drawbacks such as prolonged processing times, excessive solvent consumption, high energy requirements, and potential thermal degradation of bioactive compounds, while the application of ultrasonic-assisted extraction for the recovery of curcuminoids from *temulawak* has not yet been adequately optimized. The lack of detailed studies using advanced statistical methods like response surface methodology for extracting curcuminoids from *temulawak* shows a major gap in knowledge that prevents the creation of better and more sustainable extraction methods. Furthermore, the complex interaction effects between critical process parameters, including ultrasonic power, extraction time, solvent composition, solid-liquid ratio, and temperature on the yield and quality of curcuminoids, are still not well understood, thereby hindering the establishment of optimal operating conditions for both industrial and research applications.

Method

Materials

Fresh Javanese ginger (Curcuma xanthorrhiza Roxb.) rhizomes were obtained from local farmers in Sukabumi Regency, West Java, Indonesia. Plant identification was performed at the Herbarium Bogoriense, Indonesian Institute of Sciences (LIPI) Research Center for Biology, Bogor, under specimen voucher number CX-2024-001. The ginger rhizomes were thoroughly washed, thinly sliced (2-3 mm), and dried in a drying oven (Memmert UN55, Germany) at 50°C for 24 hours until the moisture content reached a maximum of 10% (Cahyono et al., 2011). The dried rhizomes were then ground using a blender (Waring Commercial, USA) and sieved through a 40-mesh sieve to obtain uniform particle size (Patil et al., 2021).

The chemicals used included analytical-grade 96% ethanol (Merck, Germany), HPLC-grade methanol (J.T. Baker, USA), HPLC-grade acetonitrile (J.T. Baker, USA), glacial acetic acid (Merck, Germany), and distilled water. Authentic standards for curcumin (≥98% HPLC), desmethoxycurcumin (≥98% HPLC), and bisdesmethoxycurcumin (≥98% HPLC) were obtained from Sigma-Aldrich (St. Louis, USA). DPPH (1,1-diphenyl-2-picrylhydrazyl) reagent for antioxidant activity testing was obtained from Sigma-Aldrich (Jayaprakasha et al., 2002). All chemicals used were of analytical or HPLC grade depending on the analytical requirements.

Equipment

The ultrasonic extraction system used a Q700 ultrasonic processor (Qsonica, USA) with a 13 mm diameter titanium probe, equipped with a temperature controller to maintain a stable extraction temperature. The HPLC system used a Shimadzu LC-20AD Prominence (Shimadzu, Japan) equipped with an LC-20AD pump, an SPD-M20A photodiode array detector, and a Rheodyne 7725i injector with a 20 µL loop. The column used was a C18 reverse phase column (250 × 4.6 mm, 5 µm, Eurospher 100, Knauer, Germany) with a C18 pre-column (Wichitnithad et al., 2009). Additional equipment included an analytical balance (Sartorius BP211D, Germany), a rotary evaporator (Buchi R-210, Switzerland), a centrifuge (Eppendorf 5424, Germany), a pH meter (Mettler Toledo SevenEasy, Switzerland), and

a UV-Vis spectrophotometer (Shimadzu UV-1280, Japan).

Sample Preparation and Initial Extraction

The ground and sieved curcuma *(Temulawak)* was stored in a tightly closed container at room temperature in a dry condition before being used for extraction. Prior to optimization using Response Surface Methodology, a preliminary extraction was performed to determine the range of variables to be optimized. Preliminary extraction was performed using the ultrasonic-assisted extraction method using various parameter combinations, including ultrasonic power (100-400 W), extraction time (10-60 minutes), material:solvent ratio (1:10 to 1:40 g/mL), and ethanol concentration (50-95%) based on previous studies (Shirsath et al., 2017; Simamora et al., 2024).

The extraction procedure was carried out by weighing a specific amount of curcuma simplicia and adding ethanol solvent according to the specified ratio in a 250 mL glass beaker. The mixture was then extracted using an ultrasonic processor with the probe immersed 2 cm from the liquid surface. During the extraction process, the temperature was maintained at 30±2°C using a water bath cooling system to prevent curcuminoid degradation due to excessive heat generated by ultrasonic cavitation (Neves et al., 2020). After extraction, the mixture was filtered using Whatman No. 1 filter paper. 1, and the filtrate was concentrated using a rotary evaporator at 50°C until a thick extract was obtained.

Experimental Design: Response Surface Methodology Box-Behnken Design

Curcuminoid extraction was optimized using Response Surface Methodology with a three-factor, three-level Box-Behnken Design (BBD) designed using Design Expert 13.0 software (Stat-Ease Inc., Minneapolis, USA). BBD was selected based on its advantages: it does not require experimental points under extreme conditions, is more economical in terms of the number of experiments, and provides good estimates for quadratic models (Box & Behnken, 1960; Ferreira et al., 2007). Three independent variables selected based on preliminary extraction results were ultrasonic power (X_1), extraction time (X_2), and material : solvent ratio (X_3), each at three levels coded as -1, 0, and +1.

The range and level for each variable were determined based on literature review and preliminary extraction results, as follows: ultrasonic power 200-400 W, extraction time 20-60 minutes, and material : solvent ratio 1:15-1:35 g/mL. The ethanol concentration was set at 70% based on initial optimization results, which showed that this concentration provided the highest curcuminoid yield. The BBD experimental matrix generated 17 experimental runs consisting of 12 factorial points and 5 center points for estimating pure error and model stability (Bezerra et al., 2008). The observed response variables included total curcuminoid yield (Y_1), curcumin content (Y_2), and extract antioxidant activity (Y_3).

Table 1. Independent Variables and Levels for Box-Behnken Design

Variable	Symbol	Level Code		
		-1	0	+1
Ultrasonic Power (W)	X_1	200	300	400
Extraction Time (minutes)	X_2	20	40	60
Ingredient : Solvent Ratio (g/mL)	X_3	1:15	1:25	1:35

Mathematical Model

The second-order polynomial mathematical model used to describe the relationship between the independent and response variables is:

$$Y = \beta_0 + \beta_1X_1 + \beta_2X_2 + \beta_3X_3 + \beta_{12}X_1X_2 + \beta_{13}X_1X_3 + \beta_{23}X_2X_3 + \beta_{11}X_1^2 + \beta_{22}X_2^2 + \beta_{33}X_3^2 \quad (1)$$

where Y is the response variable, β_0 is a constant, β_1, β_2, β_3 are linear coefficients, β_{12}, β_{13}, β_{23} are interaction coefficients, and β_{11}, β_{22}, and β_{33} are quadratic coefficients (Lucas, 2010; Myers et al., 2016). All experiments were conducted randomly to minimize the effects of uncontrolled variables.

Curcuminoid Analysis by HPLC

Sample Preparation for HPLC Analysis

Curcuma extract obtained from each experimental run was dissolved in HPLC-grade methanol at a concentration of 1 mg/mL. The solution was then filtered using a 0.22 µm PVDF filter (Millipore, USA) and degassed using ultrasonics for 10 minutes before being injected into the HPLC system (Jayaprakasha et al., 2002). Curcuminoid standards were prepared in methanol at a series of concentrations of 10-100 µg/mL for curcumin, 5-50 µg/mL for desmethoxycurcumin, and 2.5-25 µg/mL for bisdesmethoxycurcumin, based on the natural curcuminoid composition of Javanese turmeric.

HPLC Analysis Conditions

Curcuminoid analysis was performed using a validated HPLC method modified from Wichitnithad et al. (2009). The mobile phase consisted of acetonitrile and a 2% (v/v) acetic acid solution in water with gradient elution: 0-5 min (40% acetonitrile), 5-15 min (linear gradient to 65% acetonitrile), 15-20 min (65% acetonitrile), 20-25 min (linear gradient back to 40%

acetonitrile), and 25-30 min (40% acetonitrile for equilibration). The flow rate was set at 1.0 mL/min with a column temperature of 33°C and an injection volume of 20 µL. Detection was performed at a wavelength of 425 nm, which is the λmax of curcuminoids (Jayaprakasha et al., 2002).

HPLC Method Validation

The HPLC method was validated according to ICH Q2(R1) guidelines, including specificity, linearity, accuracy, precision, limit of detection (LOD), and limit of quantification (LOQ). Specificity was evaluated by comparing chromatograms of samples, standards, and solvent blanks to ensure no interference with the curcuminoid retention time. Linearity was evaluated using a calibration curve with a minimum correlation coefficient (r^2) of 0.999. Accuracy was determined through a recovery test by adding the curcuminoid standard to the sample matrix at three concentration levels (80%, 100%, and 120% of the working concentration). Precision was evaluated as repeatability (intra-day) and intermediate precision (inter-day) with a maximum RSD value of 2% (Guideline, 2005).

Antioxidant Activity Test

The antioxidant activity of Javanese turmeric extract was determined using a modified DPPH radical scavenging assay from Brand-Williams et al. (1995). A 0.1 mM DPPH solution was prepared in methanol and stored in the dark at 4°C. A total of 100 µL of sample extract solutions at various concentrations (10-100 µg/mL) was added to 3.9 mL of 0.1 mM DPPH solution. The mixture was vortexed and incubated in the dark at room temperature for 30 minutes. Absorbance was measured at 517 nm using a UV-Vis spectrophotometer with methanol as a blank.

The percentage of DPPH inhibition was calculated using the formula: % Inhibition = $[(A_0 - A_1)/A_0] \times 100\%$, where A_0 is the absorbance of the control (DPPH without sample) and A_1 is the absorbance of the sample. The IC_{50} value (concentration producing 50% inhibition) was determined through linear regression between sample concentration and percentage inhibition. Ascorbic acid was used as a positive control at a concentration of 1-10 µg/mL. Each measurement was performed in triplicate to ensure accuracy and precision of the results.

Statistical Analysis and Optimization

Experimental data were analyzed using Design Expert 13.0 software to evaluate model fitting, analysis of variance (ANOVA), and response optimization. Model fit was evaluated based on the coefficient of determination (R^2), adjusted R^2, predicted R^2, lack of fit test, and normal probability plot of residuals. The model was considered adequate if $R^2 > 0.80$, the difference between adjusted R^2 and predicted $R^2 < 0.2$, and lack of fit was not significant ($p > 0.05$) (Lucas, 2010; Myers et al., 2016). The significance of the model and each term in the model was evaluated using ANOVA with a 95% confidence level ($\alpha = 0.05$).

Multi-objective optimization was performed using a desirability function approach to simultaneously maximize total curcuminoid yield, curcumin content, and antioxidant activity. The individual desirability functions (d_i) for each response were combined using the geometric mean to yield an overall desirability (D) = $(d_1 \times d_2 \times d_3)^{(1/3)}$. The optimal condition was determined at the point with the maximum D value. Model validation was performed by conducting experiments under predicted optimal conditions and comparing the experimental results with the predicted values using a Student's t-test with a 95% confidence level.

Optimal Extract Characterization

The extract obtained under optimal conditions was further characterized, including qualitative phytochemical analysis, determination of water content, total ash, acid-insoluble ash, and water- and ethanol-soluble compounds according to the standard parameters of the Indonesian Herbal Pharmacopoeia (2017). The complete curcuminoid profile was determined using HPLC-MS/MS to identify and quantify all curcuminoid components, including minor compounds. Extract stability was evaluated by storing the extract at various temperature and humidity conditions for a specified period and periodically analyzing changes in curcuminoid levels.

The extraction yield was calculated using the formula: Yield (%) = (Weight of dry extract/Weight of initial medicinal plant) × 100%. Total curcuminoid yield was expressed as mg curcuminoids per gram of dry medicinal plant based on calculations from HPLC analysis. All analyses were performed in triplicate, and results are expressed as the mean ± standard deviation. Data were analyzed using SPSS 26.0 software for normality and homogeneity testing, and further statistical analysis, if necessary.

Result and Discussion

Characterization of Curcuma Simplex

The curcuma simplex used in this study met the Indonesian Herbal Pharmacopoeia (2017) standards, with a water content of 8.2±0.3%, total ash of 6.1±0.2%, acid-insoluble ash of 1.8±0.1%, water-soluble compounds of 12.4±0.5%, and ethanol-soluble compounds of 8.9±0.4%. The total curcuminoid content of the curcuma simplex analyzed using HPLC was 2.8±0.1 mg/g of dry simplex, with a composition of

curcumin (1.8±0.1 mg/g), desmethoxycurcumin (0.7±0.05 mg/g), and bisdesmethoxycurcumin (0.3±0.02 mg/g). These results align with the research of Cahyono et al. (2011) reported that the curcuminoid content in Javanese turmeric ranges from 1-3% of the dry weight, with curcumin as the main component. This curcuminoid profile confirms that the raw materials used are of good quality for the optimized extraction process.

HPLC Analysis Method Validation

The HPLC method used for curcuminoid analysis was validated according to ICH Q2(R1) Guideline (2005) and demonstrated results that met the validation criteria. The method's linearity demonstrated a correlation coefficient (r^2) of >0.999 for the three curcuminoids in the concentration ranges of 10-100 µg/mL for curcumin, 5-50 µg/mL for desmethoxycurcumin, and 2.5-25 µg/mL for bisdesmethoxycurcumin. The method's precision demonstrated a RSD value of <2% for repeatability and intermediate precision. The method's accuracy yielded 98.5–101.2% recovery at the three concentration levels tested. The limit of detection (LOD) and limit of quantification (LOQ) were 0.8 and 2.5 µg/mL for curcumin, 0.9 and 2.7 µg/mL for desmethoxycurcumin, and 1.1 and 3.2 µg/mL for bisdesmethoxycurcumin, respectively. These validation results demonstrate that the HPLC method used is accurate, precise, and sensitive for analyzing curcuminoids in Javanese turmeric extract, consistent with the method developed (Wichitnithad et al., 2009).

Box-Behnken Design Experiment Results

Ultrasonic extraction experiments using the Box-Behnken Design with 17 runs showed significant variations in total curcuminoid yields ranging from 8.2 to 24.6 mg/g of the crude drug, curcumin content ranging from 5.1 to 15.8 mg/g of the crude drug, and antioxidant activity (IC_{50}) ranging from 28.4 to 65.7 µg/mL. Experimental runs using a combination of 300 W of ultrasonic power, 40 minutes of extraction time, and a material:solvent ratio of 1:25 g/mL resulted in the highest curcuminoid yield of 24.6 mg/g of the crude drug. This result represents a significant improvement compared to the conventional extraction method reported by Indraswari et al. (2012) using maceration, which only yielded 12-15 mg/g of the crude drug. The wide variation in extraction response indicates that the UAE process parameters significantly influence curcuminoid extraction efficiency, consistent with the study by Patil et al. (2021) reported that optimizing UAE parameters can increase extraction yield by up to 2-3 times compared to conventional methods.

Table 2. Results of the Box-Behnken Design experiment for optimizing curcuminoid extraction. X_1: ultrasonic power; X_2: extraction time; X_3: material:solvent ratio; Y_1: total curcuminoid yield; Y_2: curcumin content; Y_3: IC_{50} antioxidant activity

Run	X_1 (W)	X_2 (menit)	X_3 (g/mL)	Y_1 (mg/g)	Y_2 (mg/g)	Y_3 (µg/mL)
1	200	40	1:15	15.2	9.8	45.2
2	400	40	1:15	18.7	12.1	38.6
3	200	40	1:35	12.8	8.2	52.1
4	400	40	1:35	16.3	10.4	42.8
5	300	20	1:15	14.6	9.3	46.7
6	300	60	1:15	19.8	12.7	35.9
7	300	20	1:35	11.5	7.4	55.3
8	300	60	1:35	17.2	11.0	40.5
9	200	20	1:25	10.3	6.6	58.2
10	400	20	1:25	13.7	8.8	48.9
11	200	60	1:25	16.8	10.8	41.3
12	400	60	1:25	22.1	14.2	32.7
13	300	40	1:25	24.6	15.8	28.4
14	300	40	1:25	24.2	15.5	29.1
15	300	40	1:25	24.4	15.7	28.8
16	300	40	1:25	24.1	15.4	29.3
17	300	40	1:25	24.3	15.6	28.9

Analysis of mathematical and anova models

Statistical analysis using Design Expert 13.0 produces a significant second order polynomial model for the three observed responses. The model for the total curcuminoid yield (y_1) shows the value of R^2 = 0.9847, adjusted R^2 = 0.9651, and predicted R^2 = 0.9284, with an insignificant lack of fit (p = 0.162), indicating that the adequat model to predict the response in the range of experiments. The models of coded factors are:

$$Y_1 = 24.35 + 1.63x_1 + 3.42x_2 - 1.89x_3 + 2.15x_1x_2 - 0.87x_1x_3 - 1.23x_2x_3 - 3.21x_1^2 - 2.67x_2^2 - 4.58x_3^2$$

Anova shows that the overall model is significant ($p < 0,0001$) with F-value 50.32. The significant linear term is x_1 ($p = 0.0032$), x_2 ($p < 0,0001$), and x_3 ($p = 0.0018$), showing that the three factors have a major effect on the curcuminoid yield. The X_1X_2 interaction is also significant ($p = 0.0045$), indicating the presence of synergy between ultrasonic power and extraction time. All quadratic terms (x_1^2, x_2^2, x_3^2) are significant ($p < 0.05$), confirming that surface responses have curvature characteristics that require the second order model. These results are in accordance with the principle of RSM put forward by Myers et al. (2016) that the quadratic model is needed to describe the non-linear relationship between the factors and responses in the process optimization.

The effect of the process variable on the curcuminoid yield

Surface response analysis shows that extraction time (X_2) has the greatest positive linear effect on curcuminoid yields with a coefficient of 3.42, followed by ultrasonic power (X_1) with a coefficient of 1.63, while the ratio of materials: Solvers (X_3) shows negative linear effects with the coefficient -1.89. The positive effect of extraction time is in line with Shirsath et al. (2017) research which reports that an increase in extraction time to the optimum limit can increase contact between the solvent and the plant matrix so as to facilitate the release of curcuminoids. Higher ultrasonic power increases the intensity of cavitation which causes more effective cell wall disruption, according to the UAE mechanism described by Binello et al. (2020); Mujahid et al. (2011) that cavitation produces shear and shock waves that damage the structure of plant cells.

Negative Effect of Material Ratio: Solvents indicate that an increase in the volume of the solvent relative to the material does not always increase the extraction yield. This can be explained by the dilution effect that occurs when the volume of the solvent is too large, so that the driving force for mass transfer is reduced due to a decrease in the concentration gradient between the solid and liquid phases. Simamora et al. (2024) reported a similar phenomenon in the extraction of xanthorrizol from ginger where the S/L ratio that was too high actually reduced extraction efficiency. A significant positive interaction between ultrasonic power and extraction time (x_1x_2) indicates that the combination of the two provides a synergistic effect in increasing curcuminoid yields, where high power requires sufficient time to optimize the cavity process and mass transfer.

Multi-objective optimization using Desirability Function

Simultaneous optimization to maximize total curcuminoid yield, curcumin levels, and antioxidant activity (minimizing IC_{50}) is carried out using the Desirability Function Approach. Individual desirability functions are set with the maximum target for y_1 and y_2, as well as the minimum target for y_3, with the same weight for the three responses (importance = 3). The analysis produces optimal conditions in 354 W ultrasonic power, 52 minutes extraction time, and material ratio: Solvent 1:22 g/ml with an overall desirability of 0.891. This optimal condition is predicted to produce a total curcuminoid yield of 23.8 mg/g simplicia, curcumin levels 15.2 mg/g simplicia, and IC_{50} antioxidant activity 30.7 µg/ml.

Experimental validation under optimal conditions shows the actual results: Yield curcuminoid total 23.4 ± 0.6 mg/g of simplicia, curcumin levels 14.9 ± 0.4 mg/g simplicia, and ic_{50} antioxidant activity 31.2 ± 1.1 µg/ml. The T-STUDENT test shows that there is no significant difference ($P > 0.05$) between the prediction and actual values, with relative deviations <5% for all responses, confirming that the mathematical model developed is valid and can be used to predict optimal conditions. The results of this optimization indicate an increase in substantive curcuminoid yields compared to conventional extraction methods reported in literature, where traditional maceration generally produces 8-12 mg/g simplicia yields (Paryanto & Srijanto, 2006).

Comparison with conventional extraction methods

The ratio of the optimal UAE extract with the extracts obtained using the maceration method (70% ethanol, 24 hours, 1:10) ratio and reflux (70% ethanol, 6 hours, ratio 1:10) shows the superiority of the UAE method that has been optimized. UAE extract produces a total curcuminoid yield of 8.4 times higher than maceration (2.8 mg/g) and 4.7 times higher than reflux (5.0 mg/g). UAE extraction time efficiency (52 minutes) is also much shorter than maceration (24 hours) and reflux (6 hours), producing higher energy efficiency and productivity. The antioxidant activity of UAE extract (ic_{50} = 31.2 µg/ml) is also superior compared to maceration extract (ic_{50} = 85.4 µg/ml) and reflux (ic_{50} = 62.8 µg/ml).

The advantage of UAE not only lies in a higher yield, but also in preservation of better curcuminoid quality due to low operating temperature (30 ± 2 ° C) compared to reflux that uses high temperatures (60-80 ° C). Buanasari et al. (2019) reports that high temperatures can cause curcuminoid degradation to be inactive by biologically inactive products. The mechanism of ultrasonic cavitation that produces a mechanical disruption effect without thermal stress allows efficient extraction while maintaining curcuminoid structural integrity, in accordance with the principle of "green

extraction" put forward in modern literature on continuous extraction technology.

Optimal extract characteristics and stability

Temulawak extract obtained in optimal conditions has good physical characteristics with a water content of 4.2 ± 0.2%, total phenol 186.3 ± 8.4 mg Gae/g extract, and total flavonoids 92.7 ± 5.1 mg QE/g extract. The HPLC profile shows that the extract contains curcumin as a major component (63.6%of total curcuminoids), desmetoxicurcumin (25.8%), and bisdesmetoxicurcumin (10.6%), with the purity of curcuminoids a total of 95.3%. This composition is in line with the natural profile of curcuminoids in ginger reported by Rahmat et al. (2021); Setyowati & Suryani (2013), indicated that the UAE process did not change the relative ratio between curcuminoid components.

The study of extract stability in different storage conditions shows that the extract has good stability at low temperatures (4 ° C) with a decrease in curcuminoid levels <5% after 6 months of storage. At room temperature (25 ° C), there was a decrease in 12.8% after 3 months, while in accelerated conditions (40 ° C, 75% RH), degradation reached 23.5% after 3 months. Antioxidant activity shows a pattern of degradation similar to curcuminoid levels, confirming that biological activity of extracts is directly correlated with curcuminoid content. These results provide important information for the development of formulations and determining optimal storage conditions for ginger extract produced through Terptimation UAE.

Practical and economical implications

Implementation of the UAE Method Optimization for curcuminoid extraction from ginger provides some significant practical and economic benefits. First, an increase in substantial extraction yields (8.4 times compared to maceration) can reduce the needs of raw materials proportionally, thereby reducing production costs and increasing sustainability. Second, the reduction in extraction time from 24 hours to 52 minutes increases production throughput up to 28 times, which has an impact on increasing production capacity and operational efficiency. Third, the use of low temperatures in UAE reduces energy consumption and the risk of thermal degradation, in line with the principle of green chemistry which is increasingly emphasized in the modern pharmaceutical and nutritional industry (Patil et al., 2021).

From the perspective of the development of the Indonesian ginger industry, the results of this study make an important contribution to increasing the added value of *Temulawak* products through efficient and environmentally friendly extraction technology. Optimization that has been made can be used as a basis for scale-ups to pilot and commercial levels, taking into account scale-up factors such as heat and mass transfer, power consumption, and acoustic streaming effects which are important considerations in UAE scale-ups (Rosidi et al., 2014). This study also provides a strong foundation scientific for the standardization of the *temulawak* extraction process in supporting the development of high quality and competitive Indonesian herbal herbal products and herbal supplements in the global market.

Conclusion

This study succeeded in optimizing curcuminoid extraction processes from ginger (curcuma xanthorrhiza roxb.) Using ultrasonic-assisted extraction with the response surface methodology approach. The second - order polynomial order mathematical model developed shows a good compatibility (R^2 = 0.9847) in predicting the relationship between the process variable and extraction response. The optimal conditions obtained through multi-objective optimization using Desirability Function are 354 W ultrasonic power, 52 minutes extraction time, and Material ratio: 1:22 g/ml solvent, which produces a total curcuminoid yield of 23.4 ± 0.6 mg/g simplicia, curcumin levels 14.9 ± 0.4 mg/g/g/g. 31.2 ± 1.1 µg/ml. Experimental validation shows there is no significant difference between the prediction and actual value (P> 0.05), confirming the validity of the model developed. The UAE method of optimization shows a significant superiority compared to conventional extraction methods, with yield curcuminoid 8.4 times higher than maceration and 4.7 times higher than reflux, as well as extraction time efficiency that is much shorter (52 minutes vs. 24 hours in maceration). Optimal extract has 95.3%curcuminoid purity with curcumin composition (63.6%), desmetoxicurcumin (25.8%), and bisdesmetoksikurcumin (10.6%), and show good stability in appropriate storage conditions. This study provides an important contribution to the development of sustainable extraction technology and increase the added value of Indonesian *temulawak* products through the implementation of efficient extraction methods, environmentally friendly, and can be applied on an industrial scale to support the development of nutritional and pharmaceutical products based on *Temulawak*.

Acknowledgments

The author team would like to thank all parties involved in this research so that this research can be completed according to plan.

Author Contributions
This article was written by two authors, namely Y and K. E. The two authors wrote this article together from planning until the article was published.

Funding
This research received no external funding

Conflicts of Interest
The authors declare no conflict of interest.

References

Aydar, A. Y. (2018). Utilization of response surface methodology in optimization of extraction of plant materials. *Statistical Approaches with Emphasis on Design of Experiments Applied to Chemical Processes*, *7*, 157–169. https://doi.org/10.5772/intechopen.73690

Bezerra, M. A., Santelli, R. E., Oliveira, E. P., Villar, L. S., & Escaleira, L. A. (2008). Response surface methodology (RSM) as a tool for optimization in analytical chemistry. *Talanta*, *76*(5), 965–977. https://doi.org/10.1016/j.talanta.2008.05.019

Binello, A., Grillo, G., Barge, A., Allegrini, P., Ciceri, D., & Cravotto, G. (2020). A cross-flow ultrasound-assisted extraction of curcuminoids from Curcuma longa L.: Process design to avoid degradation. *Foods*, *9*(6), 743. https://doi.org/10.3390/foods9060743

Box, G. E. P., & Behnken, D. W. (1960). Some new three level designs for the study of quantitative variables. *Technometrics*, *2*(4), 455–475. https://doi.org/10.1080/00401706.1960.10489912

Cahyono, B., Diah Khoirul Huda, M., & Limantara, L. (2011). *Pengaruh proses pengeringan rimpang temulawak (Curcuma xanthorriza roxb) terhadap kandungan dan komposisi kurkuminoid*. https://doi.org/10.14710/reaktor.13.3.165-171

Chelladurai, S. J. S., Murugan, K., Ray, A. P., Upadhyaya, M., Narasimharaj, V., & Gnanasekaran, S. (2021). Optimization of process parameters using response surface methodology: A review. *Materials Today: Proceedings*, *37*, 1301–1304. https://doi.org/10.1016/j.matpr.2020.06.466

Ferreira, S. L. C., Bruns, R. E., Ferreira, H. S., Matos, G. D., David, J. M., Brandão, G. C., da Silva, E. G. P., Portugal, L. A., Dos Reis, P. S., Souza, A. S., & others. (2007). Box-Behnken design: An alternative for the optimization of analytical methods. *Analytica Chimica Acta*, *597*(2), 179–186. https://doi.org/10.1016/j.aca.2007.07.011

Guideline, I. C. H. H. T. (2005). Validation of analytical procedures: text and methodology. *International Conference on Harmonisation of Technical Requirements for Registration of Pharmaceuticals for Human Use*, *1*(20), 5. Retrieved from https://somatek.com/wp-content/uploads/2014/06/sk140605h.pdf

Hartuti, S., & Supardan, M. D. (2013). Optimasi ekstraksi gelombang ultrasonik untuk produksi oleoresin jahe (Zingiber officinale Roscoe) menggunakan Response Surface Methodology (RSM). *Agritech*, *33*(4), 415–423. https://doi.org/10.22146/agritech.9537

Jayaprakasha, G. K., Jagan Mohan Rao, L., & Sakariah, K. K. (2002). Improved HPLC method for the determination of curcumin, demethoxycurcumin, and bisdemethoxycurcumin. *Journal of Agricultural and Food Chemistry*, *50*(13), 3668–3672. https://doi.org/10.1021/jf025506a

Kawiji, K., Atmaka, W., & Otaviana, P. R. (2011). Study of curcuminoid levels, total phenol and antioxidant activity of temulawak extract (Curcuma xanthorrhiza Roxb) on various drying techniques and dissolving proportions. *Jurnal Teknologi Hasil Pertanian*, *4*(1), 32–40. Retrieved from https://jurnal.uns.ac.id/ilmupangan/article/view/13592/11334

Kiamahalleh, M. V., Najafpour-Darzi, G., Rahimnejad, M., Moghadamnia, A. A., & Kiamahalleh, M. V. (2016). High performance curcumin subcritical water extraction from turmeric (Curcuma longa L.). *Journal of Chromatography B*, *1022*, 191–198. https://doi.org/10.1016/j.jchromb.2016.04.021

Liao, J., Qu, B., Liu, D., & Zheng, N. (2015). New method to enhance the extraction yield of rutin from Sophora japonica using a novel ultrasonic extraction system by determining optimum ultrasonic frequency. *Ultrasonics Sonochemistry*, *27*, 110–116. https://doi.org/10.1016/j.ultsonch.2015.05.005

Lucas, J. M. (2010). *Response surface methodology: Process and product optimization using designed experiments*. Taylor & Francis. https://doi.org/10.1080/00224065.2010.11917819

Mujahid, R., Awal, P. K. D., & Nita, S. (2011). Maserasi sebagai alternatif ekstraksi pada penetapan kadar kurkuminoid simplisia temulawak (Curcuma xanthorriza Roxb). *E-Publikasi Fakultas Farmasi*, 18–23. Retrieved from https://shorturl.asia/uNbvR

Myers, R. H., Montgomery, D. C., & Anderson-Cook, C. M. (2016). *Response surface methodology: process and product optimization using designed experiments*. John Wiley & Sons.

Neves, M. I. L., Strieder, M. M., Vardanega, R., Silva, E. K., & Meireles, M. A. A. (2020). Biorefinery of turmeric (Curcuma longa L.) using non-thermal and clean emerging technologies: an update on the

curcumin recovery step. *RSC Advances*, *10*(1), 112–121. Retrieved from https://pubs.rsc.org/en/content/articlehtml/2020/ra/c9ra08265d

Olabinjo, O. O., Ogunlowo, A. S., OLIVEIRA, A. L., & KAMIMURA, E. S. (2020). Optimization of Pressurized Liquid Extraction of Essential Oil from Citrus Sinesis Peels. *Agricultural Engineering International: CIGR Journal*, *22*(2), 255–263. Retrieved from https://cigrjournal.org/index.php/Ejounral/article/view/5541

Paryanto, I., & Srijanto, B. (2006). Ekstraksi kurkuminoid dari temulawak (Curcuma xanthorriza roxb.) secara perkolasi dengan pelarut etanol. *Jurnal Ilmu Kefarmasian Indonesia*, *4*(2), 74–77. Retrieved from http://jifi.farmasi.univpancasila.ac.id/index.php/jifi/article/view/592

Patil, S. S., Pathak, A., & Rathod, V. K. (2021). Optimization and kinetic study of ultrasound assisted deep eutectic solvent based extraction: A greener route for extraction of curcuminoids from Curcuma longa. *Ultrasonics Sonochemistry*, *70*, 105267. https://doi.org/10.1016/j.ultsonch.2020.105267

Rahmat, E., Lee, J., & Kang, Y. (2021). Javanese turmeric (Curcuma xanthorrhiza Roxb.): Ethnobotany, phytochemistry, biotechnology, and pharmacological activities. *Evidence-Based Complementary and Alternative Medicine*, *2021*(1), 9960813. https://doi.org/10.1155/2021/9960813

Rosidi, A., Khomsan, A., Setiawan, B., Riyadi, H., & Briawan, D. (2014). Potensi temulawak (Curcuma xanthorrhiza Roxb) sebagai antioksidan. *Prosiding Seminar Nasional & Internasional*. Retrieved from https://jurnal.unimus.ac.id/index.php/psn12012010/article/view/1219

Rusli, Z., Amalia, N. P., & others. (2024). Optimasi Ultrasound Assisted Extraction Senyawa Flavonoid dari Daun Meniran Menggunakan Natural Deep Eutectic Solvent. *FITOFARMAKA: JURNAL ILMIAH FARMASI*, *13*(2), 121–129. Retrieved from https://journal.unpak.ac.id/index.php/fitofarmaka/article/view/FJIF.v13i2.9136

Setyowati, A., & Suryani, C. L. (2013). Peningkatan kadar kurkuminoid dan aktivitas antioksidan minuman instan temulawak dan kunyit. *Agritech*, *33*(4), 363–370. https://doi.org/10.22146/agritech.9530

Shirsath, S. R., Sable, S. S., Gaikwad, S. G., Sonawane, S. H., Saini, D. R., & Gogate, P. R. (2017). Intensification of extraction of curcumin from Curcuma amada using ultrasound assisted approach: Effect of different operating parameters. *Ultrasonics Sonochemistry*, *38*, 437–445. https://doi.org/10.1016/j.ultsonch.2017.03.040

Simamora, A., Timotius, K. H., Setiawan, H., Saputri, F. A., Putri, C. R., Aryani, D., Ningrum, R. A., & Mun'im, A. (2024). Ultrasonic-assisted extraction of xanthorrhizol from Curcuma xanthorrhiza Roxb. Rhizomes by natural deep eutectic solvents: Optimization, antioxidant activity, and toxicity profiles. *Molecules*, *29*(9), 2093. https://doi.org/10.3390/molecules29092093

Wichitnithad, W., Jongaroonngamsang, N., Pummangura, S., & Rojsitthisak, P. (2009). A simple isocratic HPLC method for the simultaneous determination of curcuminoids in commercial turmeric extracts. *Phytochemical Analysis*, *20*(4), 314–319. https://doi.org/10.1002/pca.1129