



## Microwave-Assisted Extraction of Phytochemicals in Cogongrass (*Imperata cylindrica*) Rhizome

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### Abstract

Microwave irradiation has received a lot of attention. It is a feasible green solvent extraction method as it utilizes water or alcohols at elevated temperature and controlled pressure conditions. Microwave extraction emerges as a promising technique from an economical point of view, being inexpensive, simple and efficient. Cogongrass (*Imperata cylindrica*) rhizome is a good source of antioxidants such as phenolic, flavonoid and saponin. The rhizome-root part serves as traditional medicine for the treatment of a wide range of infections. In our research, different parameters such as power of microwave (50, 75, 100, 125, 150 W), frequency of microwave (15, 20, 25, 30, 35 GHz), duration of the microwave irradiation (1.5, 2.0, 2.5, 3.0, 2.5 min), ratio of liquid to solid (water: material, 1.0:1.0, 1.5:1.0, 2.0:1.0, 2.5:1.0, 3.0:1.0), extraction temperature (70, 75, 80, 85, 90°C), number of extraction cycles (1, 2, 3, 4, 5) affecting to the microwave-assisted extraction of phytochemicals in cogongrass (*Imperata cylindrica*) rhizome were examined. Total total phenolic (mg GAE/ 100g), total flavonoid (mg GE/ 100g), saponin (%) were important indicators to identify the optimal variable. Our results showed that microwave power 100 W, frequency 30 GHz, duration 2.5 min, liquid to solid ratio 2.0:1.0, temperature 75°C, 3 cycles of extraction were appropriate for extraction of phytochemical components inside cogongrass (*Imperata cylindrica*) rhizome.

**Keywords:** Cogongrass, *Imperata cylindrica*, Rhizome, Microwave, Extraction, Phenolic, Flavonoid, saponin.

### Introduction

Micro-extracted extraction (MAE) is a method that utilizes a solvent to extract the phytochemical components from herbs. In this process, the improved extraction happens as a result of change in the herbal cell texture created by electromagnetic waves [1]. Microwaves are electromagnetic radiations having frequency from 0.3 to 300 GHz. Microwave energy is transferred directly to the herbal tissue through molecular interaction the electromagnetic field via conversions of electromagnetic energy into thermal energy [2, 3]. The combinations of thermal and mass gradients contribute to the high yield and short extraction duration of micro-extracted extraction. The variables that affect the extraction recovery of this extraction process

are the power and frequency of the microwaves, the duration of the microwave irradiation, the moisture content and particle sizes of the herbs, the kind and concentration of the solvent, the ratio of solid to liquid, the extraction temperature, the extraction pressure, and the number of extraction cycles [2]. Cogongrass (*Imperata cylindrica*) is a tufted, perennial grass with hard, creeping roots. It is used extensively for the treatment of various ailments viz. urinary calculi, retention of urine, diabetes, cardiac disorder, gout, common cough and cold, inflammation, anaemia, dermal wound, ringworm, cholera, dysentery and diarrhoea etc [4, 5]. The roots maybe crushed, juiced, or dried. Cogongrass contains tannin, saponin, flavonoid, alkaloid, and terpenoid [6, 7].

Phenolic derivatives and flavonoids natural compounds in plants and play several roles in the plant's life such as general growth, reproduction, and defence against parasites and pests. Flavonoids themselves are a group of hydroxylated phenolic compounds having a benzo- $\gamma$ -pyrone structure and are ubiquitously occurring in plants [8].

Saponins are steroid or triterpenoid glycosides, common in a large number of herbs. According to the structure of the aglycone or sapogenin, saponins are classified as neutral and acid type, the so-called neutral saponins are derivatives of steroids with spiroketal side chains which are almost exclusively present in the monocotyledonous angiosperms and the acid saponins that possess triterpenoid structure type, which is the most common and occur mainly in the dicotyledonous angiosperms. It has effect on cold blooded animals, also to have the analgesic, anti-nociceptive, antioxidant activity, to impair the digestion of protein, to cause hypoglycemia and to act as antifungal and antiviral agents [9].

Microwave-assisted extraction (MAE) has been recognized as a technique with several advantages over other extraction methods, such as reduction of costs, extraction time, energy consumption, and CO<sub>2</sub> emissions. MAE was performed to obtain essential oils from two different herbs (Gabriel Abraham Cardoso-Ugarte et al., 2013). However, there was not any research mentioned to the application of microwave-assisted extraction to extract functional components inside Cogongrass (*Imperata cylindrica*) rhizome. Objective of our study focused on different parameters such as power of microwave, frequency of microwave, duration of the microwave irradiation, ratio of liquid to solid, extraction temperature, number of extraction cycles affecting to the microwave-assisted extraction of phytochemicals in cogongrass (*Imperata cylindrica*) rhizome.

## Materials and Method

### Material

Cogongrass (*Imperata cylindrica*) rhizomes were naturally collected from Bac Lieu province, Vietnam. After collecting, they must be conveyed to laboratory for experiments. They were subjected to washing and treatment. These rhizomes were treated by different parameters such as power of

microwave (50, 75, 100, 125, 150 W), frequency of microwave (15, 20, 25, 30, 35 GHz), duration of the microwave irradiation (1.5, 2.0, 2.5, 3.0, 2.5 min), ratio of liquid to solid (water: material, 1.0:1.0, 1.5:1.0, 2.0:1.0, 2.5:1.0, 3.0:1.0), extraction temperature (70, 75, 80, 85, 90°C), number of extraction cycles (1, 2, 3, 4, 5). At the end each treatment, samples were analyzed total phenolic (mg GAE/ 100g), flavonoid (mg GE/ 100g), saponin (%) content to demonstrate the optimal variable.

## Researching Procedure

### Effect of Microwave Power (W) in Phytochemical Extraction of *Imperata Cylindrica* Root

Raw *Imperata cylindrica* root was extracted by different microwave power values (50, 75, 100, 125, 150 W). Total phenolic (mg GAE/100g), total flavonoid (mg GE/ 100g), total saponin (%) were important indicators so they were chosen to identify the optimal microwave power.

### Effect of Microwave Frequency (GHz) in Phytochemical Extraction of *Imperata cylindrica* Root

Raw *Imperata cylindrica* root was extracted by different microwave frequency values (15, 20, 25, 30, 35 GHz). Total phenolic (mg GAE/100g), total flavonoid (mg GE/ 100g), total saponin (%) were important indicators so they were chosen to identify the optimal microwave frequency.

### Effect of Microwave Irradiation Duration (min) in Phytochemical Extraction of *Imperata Cylindrica* Root

Raw *Imperata cylindrica* root was extracted by different microwave irradiation duration values (1.5, 2.0, 2.5, 3.0, 2.5 min). Total phenolic (mg GAE/100g), total flavonoid (mg GE/ 100g), total saponin (%) were important indicators so they were chosen to identify the optimal microwave irradiation duration.

### Effect of Ratio of Liquid to Solid (Water: *Imperata Cylindrica* Root) in Phytochemical Extraction

Raw *Imperata cylindrica* root was extracted by different ratio of liquid to solid values (water: *Imperata cylindrica* root, 1.0:1.0, 1.5:1.0, 2.0:1.0, 2.5:1.0, 3.0:1.0). Total phenolic (mg GAE/100g), total flavonoid (mg GE/ 100g), total saponin (%) were important

indicators so they were chosen to identify the optimal liquid to solid ratio.

### Effect of Extraction Temperature (°C) in Phytochemical Extraction of *Imperata Cylindrica* Root

Raw *Imperata cylindrica* root was extracted by different extraction temperature values (70, 75, 80, 85, 90°C). Total phenolic (mg GAE/100g), total flavonoid (mg GE/ 100g), total saponin (%) were important indicators so they were chosen to identify the optimal extraction temperature.

### Effect of the Number of Extraction Cycles in Phytochemical Extraction of *Imperata Cylindrica* Root

Raw *Imperata cylindrica* root was extracted by different numbers of extraction cycles (1, 2, 3, 4, 5). Total phenolic (mg GAE/100g), total flavonoid (mg GE/ 100g), total saponin (%) were important indicators so they were chosen to identify the optimal number of extraction cycles.

### Chemical and Statistical Analysis

Total polyphenol content (mg GAE/100 g) was determined by Folin Ciocalteu reagent method [10]. Aluminum chloride colorimetric method was used for flavonoids (mg QE/100 g) determination [11]. Total saponin content (%) was quantified by spectrophotometry [12]. The experiments were run in triplicate with three different lots of samples. Statistical analysis was performed by the Statgraphics Centurion XVI.

### Result & Discussion

#### Effect of Microwave Power (W) in Phytochemical Extraction of *Imperata Cylindrica* Root

Raw *Imperata cylindrica* root was extracted by different microwave power values (50, 75, 100, 125, 150 W). Total phenolic (mg GAE/100g), total flavonoid (mg GE/ 100g), total saponin (%) were important indicators so they were chosen to identify the optimal microwave power.

Our result showed that the optimal microwave power should be 100 W to receive the highest content of total phenolic, flavonoid and saponin.

**Table 1: Effect of microwave power to the total phenolic, flavonoid and saponin extracted from *Imperata cylindrica* root**

Microwave power (W)	50	75	100	125	150
Total phenolic (mg GAE/ 100g)	5.84±0.02 <sup>c</sup>	6.73±0.03 <sup>b</sup>	8.12±0.00 <sup>a</sup>	7.03±0.01 <sup>ab</sup>	6.12±0.02 <sup>bc</sup>
Total flavonoid (mg GE/ 100g)	2.15±0.00 <sup>c</sup>	3.02±0.02 <sup>b</sup>	4.17±0.00 <sup>a</sup>	3.25±0.02 <sup>ab</sup>	2.78±0.03 <sup>bc</sup>
Saponin (%)	9.35±0.01 <sup>bc</sup>	11.24±0.02 <sup>ab</sup>	13.48±0.03 <sup>a</sup>	10.47±0.00 <sup>b</sup>	8.56±0.00 <sup>c</sup>

Note: the values were expressed as the mean of three repetitions; the same characters (denoted above), the difference between them was not significant ( $\alpha = 5\%$ )

In one report, variable of power had no important effects on the yield of flavonoids [13]. Lucchesi et al [14]. Studied microwave extraction of cardamom. when the glands were subjected to more severe thermal stresses and localized high pressures, pressure build-up happens within the glands which results in accelerated capacity for expansion and leading to cell rupture more rapidly than in traditional extraction.

#### Effect of Microwave Frequency (GHz) in Phytochemical Extraction of *Imperata Cylindrica* Root

Microwaves are non-ionizing electromagnetic waves of frequency between 300 MHz to 300 GHz or between wavelengths of 1 cm and 1m [15]. Raw *Imperata cylindrica* root was extracted by different microwave frequency values (15, 20, 25, 30, 35 GHz). Total phenolic (mg GAE/100g), total flavonoid (mg GE/ 100g), total saponin (%) were important indicators so they were chosen to identify the optimal microwave frequency. Our result showed that the optimal microwave frequency should be 30 GHz to receive the highest content of total phenolic, flavonoid and saponin.

**Table 2: Effect of microwave frequency (GHz) to the total phenolic, flavonoid and saponin extracted from *Imperata cylindrica* root**

Microwave frequency (GHz)	15	20	25	30	35
Total phenolic (mg GAE/ 100g)	8.12±0.00 <sup>a</sup>	8.03±0.03 <sup>ab</sup>	8.00±0.02 <sup>ab</sup>	7.96±0.01 <sup>b</sup>	7.41±0.02 <sup>c</sup>
Total flavonoid (mg GE/ 100g)	4.17±0.00 <sup>a</sup>	4.04±0.01 <sup>ab</sup>	3.95±0.03 <sup>ab</sup>	3.91±0.03 <sup>b</sup>	3.73±0.02 <sup>c</sup>
Saponin (%)	13.48±0.03 <sup>a</sup>	13.11±0.02 <sup>ab</sup>	13.02±0.01 <sup>ab</sup>	12.96±0.02 <sup>b</sup>	12.41±0.01 <sup>c</sup>

Note: the values were expressed as the mean of three repetitions; the same characters (denoted above), the difference between them was not significant ( $\alpha = 5\%$ )

### Effect of Microwave Irradiation Duration (min) in Phytochemical Extraction of *Imperata Cylindrica* Root

The factors microwave power and irradiation times influence each other to a great extent [15]. Raw *Imperata cylindrica* root was extracted by different microwave irradiation duration values (1.5, 2.0, 2.5, 3.0, 2.5 min).

**Table 3: Effect of microwave irradiation duration (min) to the total phenolic, flavonoid and saponin extracted from *Imperata cylindrica* root**

Microwave irradiation duration (min)	1.5	2.0	2.5	3.0	3.5
Total phenolic (mg GAE/ 100g)	7.96±0.01 <sup>c</sup>	8.39±0.02 <sup>b</sup>	9.45±0.00 <sup>a</sup>	8.83±0.01 <sup>ab</sup>	8.11±0.02 <sup>bc</sup>
Total flavonoid (mg GE/ 100g)	3.91±0.03 <sup>c</sup>	4.74±0.01 <sup>bc</sup>	6.12±0.04 <sup>a</sup>	5.32±0.02 <sup>ab</sup>	4.97±0.03 <sup>b</sup>
Saponin (%)	12.96±0.02 <sup>c</sup>	13.44±0.02 <sup>b</sup>	15.79±0.03 <sup>a</sup>	14.28±0.00 <sup>ab</sup>	13.17±0.01 <sup>bc</sup>

Note: the values were expressed as the mean of three repetitions; the same characters (denoted above), the difference between them was not significant ( $\alpha = 5\%$ )

The yield of analyte extracted can be enhanced with an accelerate in the extraction time; however, there is a related risk of degradation of sensitive components [16]. Chemat et al [17]. Confirmed that extraction duration in microwave assisted process was found to decrease with increase in temperature.

This decrease could be realized to the fact that with increase in temperature, the vapour pressure of water present inside the celery seeds increased leading to leaching out and evaporation of volatile oil along with moisture. Extended extraction durations increased the yield of total flavonoid, while progressively decreased flavonoids and antiradical power [18]. Celia Rodríguez-Pér et al [19]. Examined the extraction of phenolic components from *Moringa oleifera* leaves by microwave-assisted extraction. They concluded that extraction time of 20 min, 42% ethanol and 158°C were the microwave-assisted extraction optimal conditions. Total phenolic content was extracted from aromatic plants such as *Rosmarinus officinalis* using

Total phenolic (mg GAE/100g), total flavonoid (mg GE/ 100g), total saponin (%) were important indicators so they were chosen to identify the optimal microwave irradiation duration. Our result showed that the optimal irradiation duration should be 2.5 min to receive the highest content of total phenolic, flavonoid and saponin.

MAE. Compared with traditional reflux extraction, MAE reduced extraction duration, limited solvent volume and increased extraction yield of total phenolics [20].

### Effect of Ratio of Liquid to Solid (Water: *Imperata cylindrica* Root) in Phytochemical Extraction

In a higher contact surface area, the extraction efficiency rises. Similarly, finer particles permit enhanced penetration of the microwave. Fine particles may stance some technical difficulties; filtration is applied to prepare the matrix. Raw *Imperata cylindrica* root was extracted by different ratio of liquid to solid values (water: *Imperata cylindrica* root, 1.0:1.0, 1.5:1.0, 2.0:1.0, 2.5:1.0, 3.0:1.0). Total phenolic (mg GAE/100g), total flavonoid (mg GE/ 100g), total saponin (%) were important indicators so they were chosen to identify the optimal liquid to solid ratio. Our result showed that the optimal liquid to solid ratio should be 2.5:1.0 to receive the highest content of total phenolic, flavonoid and saponin.

**Table 4: Effect of liquid to solid ratio to the total phenolic, flavonoid and saponin extracted from *Imperata cylindrica* root**

Liquid to solid ratio	1.0:1.0	1.5:1.0	2.0:1.0	2.5:1.0	3.0:1.0
Total phenolic (mg GAE/ 100g)	9.45±0.00 <sup>b</sup>	10.12±0.02 <sup>ab</sup>	10.93±0.00 <sup>ab</sup>	11.15±0.03 <sup>a</sup>	11.17±0.04 <sup>a</sup>
Total flavonoid (mg GE/ 100g)	6.12±0.04 <sup>b</sup>	6.75±0.01 <sup>ab</sup>	7.03±0.03 <sup>ab</sup>	7.34±0.02 <sup>a</sup>	7.36±0.02 <sup>a</sup>
Saponin (%)	15.79±0.03 <sup>c</sup>	16.53±0.00 <sup>b</sup>	16.77±0.01 <sup>ab</sup>	16.94±0.00 <sup>a</sup>	16.97±0.03 <sup>a</sup>

Note: the values were expressed as the mean of three repetitions; the same characters (denoted above), the difference between them was not significant ( $\alpha = 5\%$ )

Solvent specification is the most vital variable because the solvent affects the absorbance of the microwave energy, as determined by the dissipation factor [21, 22]. The solvent must have an affinity for the target component and a capability to absorb microwave energy [23]. Another critical factor in microwave-assisted extraction is the

ratio of the solid herbs to the amount of solvent. The bioactive ingredients in the herbs can effectively dissolve when large amounts of solvent are used, thereby leading to improved extraction yields. Gao et al [24]. And Li et al [25]. Who applied a high solvent ratio with microwave-assisted extraction, reported consistent results.

When large quantities of solvent were utilized, the extraction yield initially increased and then decreased as the solid-to-liquid ratio decreased [26, 27, 28]. When the amount of solvent was reduced (high-solid herbal materials), microwave energy may have been absorbed and dispersed by the large amount of plant materials [24], thereby increasing the solid mass, and decreasing the surface area available for solvent to penetrate the plant materials and solubilize the target molecules [29].

### Effect of Extraction Temperature (°C) in Phytochemical Extraction of *Imperata cylindrica* Root

When the temperature of water is raised, there is a steady decrease in its permittivity, viscosity and surface tension but an increase in its diffusivity characteristics.

The increased temperature can overcome the solute–matrix interaction caused by van der Waals forces, hydrogen bonding, dipole attraction of the solutes molecules and active sites in the matrix [30]. However, thermally labile compounds are degraded at elevated temperatures. Sufficient temperature is necessary to optimize the extraction efficiency, avoid thermal degradation of the target analytes, and to supply reproducible processing conditions [31, 15].

Raw *Imperata cylindrica* root was extracted by different extraction temperature values (70, 75, 80, 85, 90°C). Total phenolic (mg GAE/100g), total flavonoid (mg GE/ 100g), total saponin (%) were important indicators so they were chosen to identify the optimal extraction temperature. Our result showed that the optimal extraction temperature should be 75°C to receive the highest content of total phenolic, flavonoid and saponin.

**Table 5: Effect of extraction temperature (°C) to the total phenolic, flavonoid and saponin extracted from *Imperata cylindrica* root**

Extraction temperature (°C)	70	75	80	85	90
Total phenolic (mg GAE/ 100g)	11.15±0.03 <sup>b</sup>	13.49±0.02 <sup>a</sup>	10.43±0.02 <sup>c</sup>	9.58±0.04 <sup>d</sup>	8.17±0.04 <sup>e</sup>
Total flavonoid (mg GE/ 100g)	7.34±0.02 <sup>b</sup>	8.78±0.04 <sup>a</sup>	6.24±0.00 <sup>c</sup>	5.11±0.03 <sup>d</sup>	4.09±0.02 <sup>e</sup>
Saponin (%)	16.94±0.00 <sup>ab</sup>	17.19±0.03 <sup>a</sup>	16.83±0.01 <sup>b</sup>	14.21±0.00 <sup>c</sup>	11.46±0.01 <sup>d</sup>

Note: the values were expressed as the mean of three repetitions; the same characters (denoted above), the difference between them was not significant ( $\alpha = 5\%$ )

High-temperature extraction can be gainful with the resulting increase in solubility. This is because higher temperature causes increased intermolecular interactions within the solvent, giving increase to higher molecular motion which rises the solubility. The accelerating temperature may cause a cellular pressure build up which may create cell rupture and opening of the cell matrix, and as a result, increased elements accessibility to be extracted into the solution (Al-Harshseh M., Kingman SW., 2004).

### Effect of the Number of Extraction Cycles in Phytochemical Extraction of *Imperata cylindrica* Root

The microwave cycle used must be carefully monitored, because microwave-assisted

extraction offers quick release of the target components in the surrounding extraction solvent and longer extraction durations could accelerate the decomposition of extracted phenolics for extended extraction under these harsh conditions [32]. Raw *Imperata cylindrica* root was extracted by different numbers of extraction cycles (1, 2, 3, 4, 5). Total phenolic (mg GAE/100g), total flavonoid (mg GE/ 100g), total saponin (%) were important indicators so they were chosen to identify the optimal number of extraction cycles.

Our result showed that the optimal number of extraction cycles should be three to receive the highest content of total phenolic, flavonoid and saponin.

**Table 6: Effect of the number of extraction cycles to the total phenolic, flavonoid and saponin extracted from *Imperata cylindrica* root**

Number of extraction cycles	1	2	3	4	5
Total phenolic (mg GAE/ 100g)	13.49±0.02 <sup>c</sup>	14.29±0.03 <sup>ab</sup>	14.78±0.02 <sup>a</sup>	14.03±0.04 <sup>b</sup>	12.38±0.03 <sup>d</sup>
Total flavonoid (mg GE/ 100g)	8.78±0.04 <sup>d</sup>	9.27±0.04 <sup>bc</sup>	10.38±0.02 <sup>a</sup>	9.46±0.02 <sup>b</sup>	9.02±0.02 <sup>c</sup>
Saponin (%)	17.19±0.03 <sup>c</sup>	18.63±0.00 <sup>b</sup>	19.48±0.03 <sup>a</sup>	17.05±0.01 <sup>d</sup>	16.09±0.00 <sup>e</sup>

Note: the values were expressed as the mean of three repetitions; the same characters (denoted above), the difference between them was not significant ( $\alpha = 5\%$ )

The outcome of stirring is related to the mass transfer manner in the solvent phase. Therefore, balance between the aqueous and vapor phases can be achieved more rapidly. The use of agitation in MAE quickens the extraction by enhancing desorption and dissolution of active compounds bound to the sample matrix. Through stirring, the disadvantages of the use of low solvent-to-solid ratio can be reduced, together with the minimization of the mass transfer barrier created by the concentrated solute in a localized region resulting from insufficient solvent. It is possible to observe the

difference between suspensions with and without stirring [33].

## Conclusion

The application of microwave extraction of active substances from plant materials results in faster heating. Electromagnetic waves are indeed absorbed selectively by media possessing a high dielectric constant resulting in more effective heating. In this research, we have successfully identified major variables affecting to the phytochemical extraction inside cogongrass (*Imperata cylindrica*) rhizome by microwave.

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