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Quantitative Profiling of Bioactive Compounds and Minerals in Zingiber officinale Extract Using HPLC-DAD and ICP-MS

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ABSTRACT:

Zingiber officinale (ginger) is widely recognized for its diverse pharmacological properties and potential applications in nutraceutical formulations. However, comprehensive characterization of its phytochemical and elemental composition is essential for evaluating its quality, safety and therapeutic potential. The present study aimed to chemically characterize a dry extract of *Z. officinale* by determining its amino acid, flavonoid and elemental profiles. Quantitative phytochemical analysis was performed using high-performance liquid chromatography coupled with diode-array detection (HPLC-DAD), while elemental composition was assessed by inductively coupled plasma mass spectrometry (ICP-MS). The analysis revealed a total amino acid content of 13.80 ± 0.47 mg/g, with histidine being the predominant amino acid (5.38 ± 0.15 mg/g). Flavonoid profiling demonstrated notable concentrations of rosavin (8.10 ± 0.24 mg/g) and dihydroquercetin (6.11 ± 0.18 mg/g). Elemental analysis indicated appreciable levels of essential macrominerals, particularly potassium and magnesium, whereas the concentrations of potentially toxic heavy metals were within the established permissible limits for pharmaceutical applications. The findings demonstrate that *Z. officinale* dry extract contains a diverse array of bioactive constituents and essential minerals, supporting its potential use as a natural ingredient in nutraceutical and pharmaceutical formulations. The characterized phytochemical and elemental profile may further contribute to the development of ginger-based preparations with antioxidant, anti-inflammatory and metabolic-support applications.

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INTRODUCTION

The growing interest in natural therapeutic agents has been driven, in part, by concerns regarding the adverse effects and limitations associated with certain synthetic pharmaceuticals [1]. Among medicinal and food plants, *Zingiber officinale Roscoe* (ginger), a perennial herbaceous species of the *Zingiber*aceae family, is widely cultivated for its aromatic and pungent rhizomes. Ginger rhizomes contain a diverse range of bioactive constituents, particularly volatile oils, gingerols and shogaols, which contribute to their antioxidant, anti-inflammatory and antimicrobial properties [1-3,24]. These biological activities have established ginger as an important raw material for pharmaceutical, cosmetic and functional food applications.

Considerable research has focused on improving the recovery of bioactive constituents from ginger through optimized extraction processes. Gonzalez-Gonzalez et al. [12] emphasized the importance of controlling extraction parameters to maximize the recovery of therapeutically relevant compounds. Ultrasonic-assisted extraction (UAE) has gained particular attention as an efficient approach for recovering bioactive compounds while maintaining their biological properties. Contreras-López et al. [11] reported effective recovery of antioxidant constituents using UAE, while Murphy & Jaiswal [11] described its application for the extraction of polyphenols with antibacterial activity. Supercritical CO_2 extraction has provided another approach for improving the recovery of valuable ginger constituents, with studies by Gani et al. [14] and Castro-Muñoz [15] reporting improved extraction performance compared with conventional methods [15].

Research on ginger grown or processed in Uzbekistan has provided information on its phytochemical composition and potential applications. Qarshieva et al. [16] reported the presence of flavonoids such as quercetin and luteolin in local ginger varieties. Ismoilov & Elchiyev [17] investigated the amino acid and mineral composition of local ginger extracts, highlighting their relevance to functional food applications. Gapparova & Xudoynazarov [18] isolated 6-gingerol using high-performance liquid chromatography (HPLC), which provide an analytical basis for its quantification. Komilov & Safarova [19] reported substantial antioxidant activity in ginger extracts and associated this activity with their phenolic constituents.

Despite these studies, ginger extraction research in Uzbekistan has largely focused on conventional procedures, such as maceration and extraction with 40-70% ethanol, with limited comprehensive quantitative characterization of the resulting extracts [20]. In particular, the combined assessment of bioactive compounds, essential minerals and potentially toxic elements using advanced analytical techniques remains insufficiently explored. This limitation restricts the development of standardized ginger extracts with well-defined chemical and safety profiles. The present study therefore investigates the extraction of *Z. officinale* using an ultrasonic-assisted extraction approach and provides a comprehensive characterization of the resulting dry extract. HPLC-DAD was employed for the quantitative determination of selected amino acids and flavonoids, while inductively coupled plasma mass spectrometry (ICP-MS) was used to characterize the mineral composition and assess the levels of potentially toxic heavy metals. The combined chemical and elemental profile provide a basis for evaluating the composition, quality and safety of *Z. officinale* dry extract for potential application in nutraceutical and pharmaceutical formulations.

MATERIALS AND METHODS

2.1. Plant Material: *Zingiber officinale Roscoe* rhizomes were obtained from a commercial farm in the Yuqori Chirchiq district, Tashkent region, Uzbekistan. The raw materials were selected according to morphological characteristics, with preference given to rhizomes having a smooth outer surface and characteristic light-yellow internal coloration, in accordance with GOST ISO 1003-2016. The collected rhizomes were thoroughly washed to remove adhering soil, dried to constant weight, finely ground and passed through a sieve to obtain a uniform particle size before extraction.

2.2. Maceration: Defatted and powdered ginger rhizomes (10 g) were placed in hermetically sealed glass containers and immersed in 200 mL of the respective extraction solvents (distilled water, 70% ethanol or 80% ethanol) to maintain a constant solid-to-solvent ratio of 1:20 (w/v). The maceration process was conducted at controlled room temperature with periodic mechanical agitation for intervals of 24, 48 and 72 h. Following each designated time interval, the crude mixtures were filtered through a Whatman No. 1 filter paper and the solvent was subsequently eliminated under reduced pressure using a rotary evaporator.

2.3. Percolation: Percolation extraction was performed using a standardized laboratory glass percolator. Pulverized and defatted ginger rhizomes (10 g) were uniformly packed into the percolation column and treated with 200 mL of the selected solvents (distilled water, 70% ethanol or 80% ethanol) at a solid-to-solvent ratio of 1:20 (w/v). The extraction was carried out

at room temperature, regulating the flow rate to allow the solvent to completely permeate the plant matrix over durations of 6, 12 and 18 h. The collected eluates were filtered and concentrated to dryness under vacuum via a rotary evaporator.

2.4. Ultrasonic-Assisted Extraction (UAE): For the ultrasonic-assisted extraction, 10 g of the defatted and powdered ginger rhizome was mixed with 200 mL of the respective solvent (distilled water, 70% ethanol, or 80% ethanol) to achieve a solid-to-solvent ratio of 1:20 (w/v). The mixture was subjected to acoustic cavitation in a digital ultrasonic bath operating at a fixed frequency of 60 kHz and a constant temperature of 25 ºC. To systematically evaluate the kinetics of extraction, the process was executed for 15, 30 and 60 min. Once completed, the extracts were immediately filtered and the liquid phases were concentrated under vacuum using a rotary evaporator.

2.5. Analysis of Bioactive Compounds and Elemental Composition

2.5.1. Determination of Free Amino Acids: Free amino acids were isolated by precipitating proteins and high-molecular-weight peptides from the extract matrix using trichloroacetic acid (TCA). Briefly, a 1 mL aliquot of the liquid sample was homogenized with 1 mL of 20% TCA and incubated at room temperature for 10 min. The mixture was then subjected to centrifugation at 8,000 rpm for 15 min. The resulting supernatant was separated, concentrated under a gentle stream of air at a reduced temperature and reconstituted in a triethylamine-acetonitrile-water (1:7:1, v/v/v) coupling mixture. This reconstitution step was performed twice to completely neutralize the acidic environment.

The generated amino acid derivatives were quantitatively analyzed using an Agilent Technologies 1200 HPLC system (Agilent Technologies, USA) equipped with a Diode-Array Detector (DAD) and a Discovery HSC18 column (75 × 4.6 mm, 5 µm). The mobile phase was composed of (A) 0.14 M CH_3COONa supplemented with 0.05% triethylamine (pH 6.4) and (B) pure acetonitrile (CH_3CN). The separation was performed at a constant flow rate of 1.2 mL/min with spectrophotometric detection set at a wavelength of 269 nm [4].

2.5.2. HPLC Analysis of Flavonoids: Flavonoid quantification was initiated by weighing 10 g of the powdered ginger sample into a 300 mL flat-bottom flask, followed by the addition of 50 mL of 70% ethanol. The mixture was refluxed for 1 h at 70–80 °C under continuous magnetic stirring, followed by an additional 2 h of agitation at room temperature to ensure exhaustive extraction. The mixture was filtered and the remaining matrix residue was re-extracted twice using 25 mL portions of 70% ethanol. The combined filtrates were quantitatively diluted to 100 mL with 70% ethanol and centrifuged at 7,000 rpm for 25 min prior to chromatographic injection [8].

Chromatographic separation was performed on the Agilent 1200 HPLC system utilizing an Eclipse XDB C18 column (250 × 4.6 mm, 5 µm; Agilent Technologies, USA). Target analytes were monitored using the DAD at wavelengths of 254 nm and 272 nm. Elution was executed via a binary gradient system comprising a phosphate buffer and analytical grade acetonitrile. The gradient profile was set as follows: 0-5 min, 95:5 (v/v); 6-12 min, 70:30 (v/v); 12–13 min, 50:50 (v/v); and 13-15 min, 95:5 (v/v). The mobile phase flow rate was maintained at 0.8 mL/min, the column oven temperature was kept constant at 30 °C and the sample injection volume was 10 µL [9].

2.5.3. Determination of Macro and Microelements (ICP-MS): The total elemental profile of the dry extract was established using Inductively Coupled Plasma Mass Spectrometry (ICP-MS). A 0.5 g portion of the dry extract was accurately weighed directly into a high-pressure Teflon autoclave vessel, to which concentrated HNO_3 and hydrogen peroxide (H_2O_2) were added. Microwave-assisted acid digestion was carried out using a Berghof MWS-3+ microwave digestion system (Berghof Products + Instruments, Germany). Following the completion of the thermal digestion program, the clear solutions were quantitatively transferred to 50 mL volumetric flasks and diluted to volume with 0.5% HNO_3. The quantitative elemental determination was conducted on a NexION 2000 ICP-MS system (Perkin-Elmer, USA). Instrument calibration was performed using a certified 29-element Multi-Element Standard Solution. High-purity argon gas (99.995%) was employed as the plasma and carrier gas to maintain optimal plasma stability.

2.6. Statistical Analysis: All extraction routines and analytical measurements were executed strictly in triplicate (n=3). The experimental data are expressed as the mean values ± standard deviation (SD). Statistical evaluations were performed using one-way analysis of variance (ANOVA) followed by appropriate post-hoc tests to determine the significance of differences between the extraction methodologies, ensuring the scientific reproducibility and reliability of the findings.

RESULTS AND DISCUSSION

3.1. Efficiency of Different Extraction Methods: The comparative analysis of three extraction methods viz. maceration, percolation and ultrasonic-assisted extraction (UAE) revealed significant variations in the yield of bioactive compounds from *Z. officinale* rhizomes.

As presented in Table-1, maceration showed the lowest efficiency compared to the other methods, likely due to limited mass transfer kinetics over the 24-72 h period. In contrast, percolation allowed for more continuous solvent-solute interaction, resulting in higher extraction yields within a shorter time frame (6-18 h). The ultrasonic-assisted extraction (UAE) demonstrated the highest extraction efficiency. The application of ultrasonic waves (60 kHz) significantly disrupted the plant cell walls, facilitating the rapid release of gingerols and other phenolic compounds into the solvent. The results indicate that using 70% and 80% ethanol as solvents consistently yielded higher concentrations of bioactive constituents compared to distilled water across all methods. Specifically, the UAE method achieved a maximum yield of 2.95 mg of bioactive compounds within 60 minutes, which is substantially higher than the yields obtained through traditional maceration and percolation processes.

Table 1.

No.	Defatted Rhizome Mass, g	Extraction Solvent	Volume of Extraction Solvent, mL.	Maceration Time, h	Content of Bioactive Compounds, mg
1	10	Distilled Water	200	24	0.14 ± 0.02
2	10	Distilled Water	200	48	0.24 ± 0.01
3	10	Distilled Water	200	72	0.37 ± 0.01
4	10	70% Ethanol	200	24	0.95 ± 0.01
5	10	70% Ethanol	200	48	1.34 ± 0.01
6	10	70% Ethanol	200	72	1.47 ± 0.02
7	10	80% Ethanol	200	24	0.95 ± 0.01
8	10	80% Ethanol	200	48	1.35 ± 0.01
9	10	80% Ethanol	200	72	1.47 ± 0.02

Note: Results are presented as mean ± SD (n=3)

3.1. Efficiency of Maceration Process: The efficiency of bioactive compound extraction from *Z. officinale* rhizomes via maceration was significantly influenced by the solvent type and extraction duration. As shown in the data, the use of distilled water yielded the lowest concentrations of bioactive compounds (0.15-0.36 mg), which confirm that while water is a highly polar solvent, it is insufficient for extracting the diverse range of phenolic compounds and gingerols present in ginger. The 70% ethanol solution demonstrated superior extraction efficiency compared to water and 80% ethanol, yielding a maximum of 1.49 mg after 72 h. This suggests that the 70% ethanol concentration provides the optimal polarity for solubilizing both polar and moderately polar bioactive components in the ginger matrix. Interestingly, increasing the ethanol concentration to 80% resulted in a slight decline in extraction yield, suggesting that exceeding the optimal solvent polarity may hinder the release of certain water-soluble compounds. Furthermore, extraction yield increased with time in all solvent systems. The most significant increase occurred within the first 48 h, while the progression from 48 to 72 h showed a slower rate, indicating that the system approached equilibrium. Thus, 70% ethanol with a 1:20 solid-to-solvent ratio and a 72-h extraction time is recommended as the optimal condition for maximizing the yield of bioactive constituents. (Table-1).

3.2. Efficiency of Percolation Process: The percolation method showed a marked improvement in extraction efficiency compared to maceration, particularly regarding processing time. As shown in Table-2, the extraction yield using distilled water remained relatively low, reaching a maximum of 0.28 mg after 18 h. This confirms the limited efficacy of water in mobilizing non-polar bioactive gingerols. In contrast, the use of 70% ethanol significantly enhanced the extraction process. The maximum yield of 2.05 mg was achieved within 18 h. This outcome demonstrates that percolation facilitates a more continuous concentration gradient between the solvent and the plant material compared to the static nature of maceration. Interestingly, the results obtained with 80% ethanol were statistically similar to those of 70% ethanol, leading to the selection of 70% ethanol as the more economical and efficient solvent for this process.

A key advantage of percolation observed in this study is the rapid attainment of extraction equilibrium. While maceration required 72 h for optimal yield, percolation achieved substantial recovery within 18 h. This suggests that the continuous solvent flow in percolation effectively prevents solvent saturation around the plant matrix, thereby enhancing mass transfer kinetics. Consequently, the optimal conditions for percolation were determined to be a 1:20 solid-to-solvent ratio using 70% ethanol over an 18 h duration.

Table 2.

No.	Defatted Rhizome Mass, g	Extraction Solvent	Volume of Extraction Solvent, mL.	Percolation Time, h	Content of Bioactive Compounds, mg
1	10	Distilled Water	200	6	0.17 ± 0.01
2	10	Distilled Water	200	12	0.19 ± 0.01
3	10	Distilled Water	200	18	0.27 ± 0.02
4	10	70% Ethanol	200	6	1.34 ± 0.01
5	10	70% Ethanol	200	12	1.54 ± 0.01
6	10	70% Ethanol	200	18	2.05 ± 0.01
7	10	80% Ethanol	200	6	1.34 ± 0.01
8	10	80% Ethanol	200	12	1.54 ± 0.01
9	10	80% Ethanol	200	18	2.05 ± 0.01

Note: Results are presented as mean ± SD (n=3)

3.3. Ultrasonic-Assisted Extraction (UAE): The extraction of bioactive compounds was performed using a digital ultrasonic-assisted extraction system. Defatted and powdered ginger rhizomes (10 g) were mixed with 200 ml of the respective solvent (distilled water, 70% ethanol, or 80% ethanol), maintaining a solid-to-solvent ratio of 1:20 (g/mL). The extraction process was carried out at specific time intervals of 15, 30 and 60 min. To ensure standardization, a parallel experimental set using 5 g of raw material with 100 ml of solvent was also conducted to verify the extraction kinetics. Following the ultrasound treatment, the mixtures were centrifuged at 7,000 rpm for 15 min. The supernatant was subsequently separated and filtered through filter paper to remove residual particulate matter. The experimental parameters, including raw material mass, solvent type and extraction duration, are summarized in Table-3. As evidenced by the experimental results, the extraction efficiency in distilled water showed a progressive increase with the duration of ultrasonic treatment. Specifically, the yield of bioactive compounds rose significantly as the extraction time was extended from 15 to 30 minutes, confirming that ultrasonic energy effectively facilitates mass transfer even in polar solvents by disrupting the plant cell matrix.

Table 3. Parameters of ultrasonic extraction, including the mass of raw material, type and volume of extraction solvent, extraction method and extraction time (60 Hz, 25°C).

No.	Defatted Rhizome Mass, g	Extraction Solvent	Volume of Extraction Solvent, ml	Ultrasound-Assisted Extraction Time, min	Extractive Yield, %	Content of Bioactive Compounds, mg
1	10	Distilled Water	200	15	4.00±1.00	1.22± 0.01
2	10	Distilled Water	200	30	5.40± 0.10	1.32± 0.01
3	10	Distilled Water	200	60	6.60± 0.10	1.57± 0.01
4	10	70% Ethanol	200	15	7.90± 0.10	1.36± 0.01
5	10	70% Ethanol	200	30	9.40± 0.10	1.72± 0.01
6	10	70% Ethanol	200	60	13.90±0.10	2.35± 0.01
7	10	80% Ethanol	200	15	7.90± 0.10	1.36± 0.01
8	10	80% Ethanol	200	30	9.40± 0.10	1.72± 0.01
9	10	80% Ethanol	200	60	13.90±0.10	2.35± 0.01

Note: Results are presented as mean ± SD (n=3)

3.4. Efficiency of Ultrasonic-Assisted Extraction (UAE): The extraction of bioactive compounds from *Zingiber officinale* rhizomes using ultrasound (60 kHz, 25 °C) revealed that both solvent polarity and treatment duration are critical determinants of extraction efficiency. Despite maintaining a constant solid-to-solvent ratio (1:20), significant variations in yield were observed

across the tested conditions (Table-3). When distilled water was used, the bioactive compound recovery increased from 1.22 ± 0.01 mg (at 15 min) to 1.57 ± 0.01 mg (at 60 min). While water demonstrated a steady extraction trend, its capacity was lower than that of the ethanolic solvents, highlighting the limited solubility of certain non-polar ginger constituents in purely aqueous media. The 70% ethanol solution showed superior extraction performance. The yield of bioactive compounds increased from 1.36 ± 0.01 mg (15 min) to 2.35 ± 0.01 mg (60 min). This efficiency is attributed to the synergistic effect of ethanol-water mixtures, which provide an optimal polarity for the simultaneous solubilization of a broader spectrum of bioactive phytochemicals. Furthermore, although 80% ethanol yielded results comparable to 70% ethanol (maximum yield of ~2.35 mg), it provided no significant advantage during shorter extraction intervals. Given these results, 70% ethanol is identified as the most suitable solvent. It combines high recovery efficiency with lower ethanol consumption, offering a cost-effective and technologically scalable solution for standardized extract production.

3.5. Characterization and Phytochemical Profiling of Dry Ginger Extract: The extraction process yielded a total dry extract of 14% (w/w) from the *Zingiber officinale* rhizomes. The obtained dry extract was evaluated for its organoleptic properties and physical appearance. The extract was characterized as a non-hygroscopic, light yellow powder, possessing a distinct pungent odor and a bitter taste, which are typical of high-quality ginger extracts. The phytochemical profiling of the dry extract, specifically focusing on the amino acid content, was conducted using HPLC-DAD. The analysis identified a diverse profile of amino acids, which confirms the nutritional and therapeutic potential of the standardized extract. The quantitative determination of these amino acids indicates that the extraction process, particularly utilizing the 70% ethanol solvent system, effectively preserves the integrity of these polar bioactive constituents. These findings validate the scalability of our optimized extraction protocol for producing consistent, high-grade ginger powder suitable for both nutraceutical and pharmaceutical applications (Table-4).

Table 4. Amino Acid Profile of *Zingiber officinale* Dry Extract

Amino Acid	Concentration (mg/g)
Aspartic acid	0.50 ± 0.02
Glutamic acid	1.42 ± 0.05
Serine	0.56 ± 0.02
Glycine	0.25 ± 0.01
Asparagine	0.49 ± 0.02
Glutamine	0.68 ± 0.03
Cysteine	1.31 ± 0.04
Threonine	0.92 ± 0.03
Arginine	0.23 ± 0.01
Alanine	0.42 ± 0.02
Proline	0.72 ± 0.03
Tyrosine	0.26 ± 0.01
Valine	0.21 ± 0.01
Methionine	0.08 ± 0.00
Histidine	5.38 ± 0.15
Isoleucine	0.11 ± 0.01
Leucine	0.11 ± 0.00
Tryptophan	0.11 ± 0.01
Phenylalanine	0.03 ± 0.00
Lysine	0.03 ± 0.00
Total	13.80 ± 0.47

Note: Data are presented as mean ± standard deviation (SD) of triplicate experiments. Values were rounded to two decimal places for clarity.

3.6. Amino Acid Profile Analysis: The amino acid composition of the dry *Z. officinale* extract was determined using HPLC-DAD. The quantitative analysis revealed a total amino acid content of 13.80 ± 0.47 mg/g (Table-4). Histidine was identified as the predominant amino acid, with a concentration of 5.38 ± 0.15 mg/g, accounting for a significant portion of the total profile. As a precursor to various metabolic pathways and a compound with known antioxidant properties, the high concentration of histidine underscores the therapeutic potential of the extract. Furthermore, significant levels of glutamic acid (1.42 ± 0.05

mg/g), cysteine (1.31 ± 0.04 mg/g) and threonine (0.92 ± 0.03 mg/g) were observed, which are essential for protein synthesis and cellular homeostasis. Other amino acids, including proline (0.72 ± 0.03 mg/g), serine (0.56 ± 0.02 mg/g), aspartic acid (0.50 ± 0.02 mg/g), asparagine (0.49 ± 0.02 mg/g) and alanine (0.42 ± 0.02 mg/g), were present in moderate concentrations, contributing to the metabolic support and tissue regeneration profile of the extract. The remaining amino acids, such as valine, tyrosine, arginine, etc. were detected in concentrations below 0.3 mg/g. Thus, the high concentration of histidine, glutamic acid and cysteine confirms the significant pharmacological and nutraceutical value of the ginger extract. The standardized profile of these bioactive constituents supports the potential application of *Z. officinale* as a potent natural source for anti-inflammatory and antioxidant formulations.

The amino acid profile of the *Z. officinale* extract is presented in Fig. 1, illustrating the concentrations of the identified amino acids. The HPLC-DAD chromatogram shown in Fig. 2 presents the separation of the detected amino acids, with distinct peaks corresponding to the identified compounds.

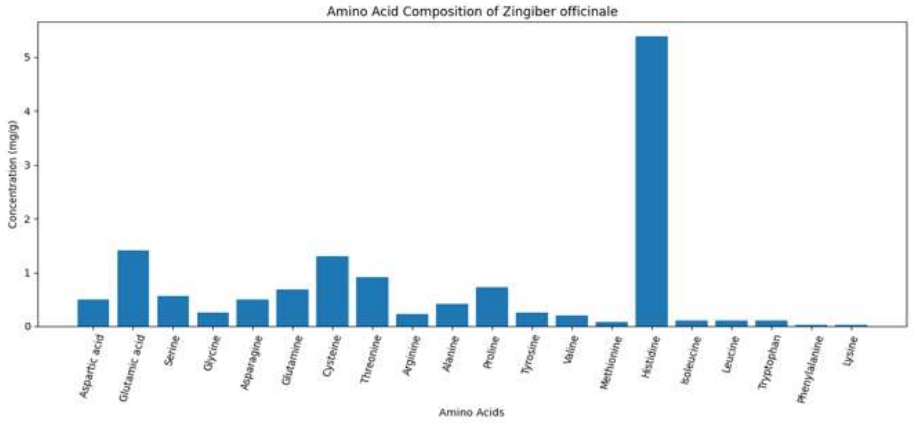


Fig. 1. Amino acid profile of *Z. officinale* dry extract (mg/g)

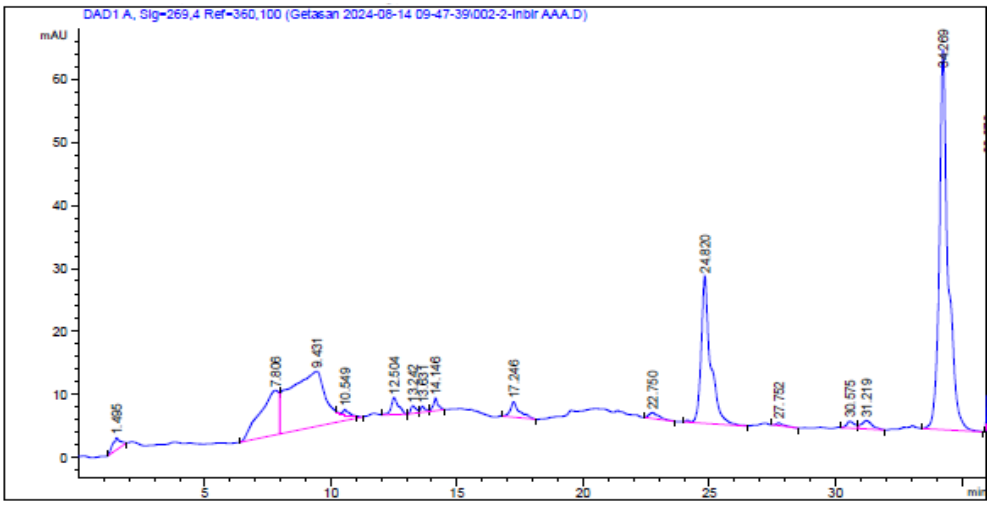


Fig. 2. HPLC-DAD chromatogram of *Z. officinale* dry extract at 269 nm

3.7. Flavonoid Profiling of Ginger Extract: The flavonoid content within the standardized *Z. officinale* extract was quantitatively determined using HPLC-DAD. This analysis is critical for assessing the antioxidant and anti-inflammatory properties associated with ginger preparations (Table-5). As shown in the analytical results, the HPLC-DAD method enabled the effective separation and identification of major flavonoid derivatives. These compounds contribute significantly to the extract's therapeutic efficacy. The phytochemical profile supports the efficiency of the optimized 70% ethanol extraction in recovering key bioactive metabolites, making it a suitable approach for nutraceutical development.

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Table 5.

Flavonoid	Concentration (mg/g)
Dihydroquercetin	6.11 ± 0.18
Rosavin	8.10 ± 0.24
Rutin	5.08 ± 0.15
Salidroside	3.17 ± 0.09
Total	22.46 ± 0.66

The quantitative determination of flavonoids in the *Z. officinale* extract is presented in Table 5. The analysis revealed a diverse flavonoid profile, with varying concentrations among the identified compounds. Rosavin was identified as the predominant flavonoid, with a concentration of 8.10 ± 0.24 mg/g. As a well-known adaptogen, the high concentration of rosavin suggests that the extract possesses significant potential in stress-modulating and adaptogenic applications. Dihydroquercetin was observed as the second most abundant component (6.11 ± 0.18 mg/g), acting as a potent antioxidant by effectively neutralizing free radicals. Furthermore, significant levels of rutin (5.08 ± 0.15 mg/g) and salidroside (3.17 ± 0.09 mg/g) were detected. Rutin is recognized for its vascular-strengthening and anti-inflammatory properties, while salidroside contributes to neuroprotective and anti-stress functions (Fig. 3). Thus, the high content of rosavin and dihydroquercetin underscores the potent antioxidant and pharmacological activity of the ginger extract. These findings confirm the functional value of *Z. officinale* as a promising raw material for nutraceutical and pharmaceutical development, emphasizing its potential in therapeutic formulations aimed at oxidative stress mitigation and metabolic support.

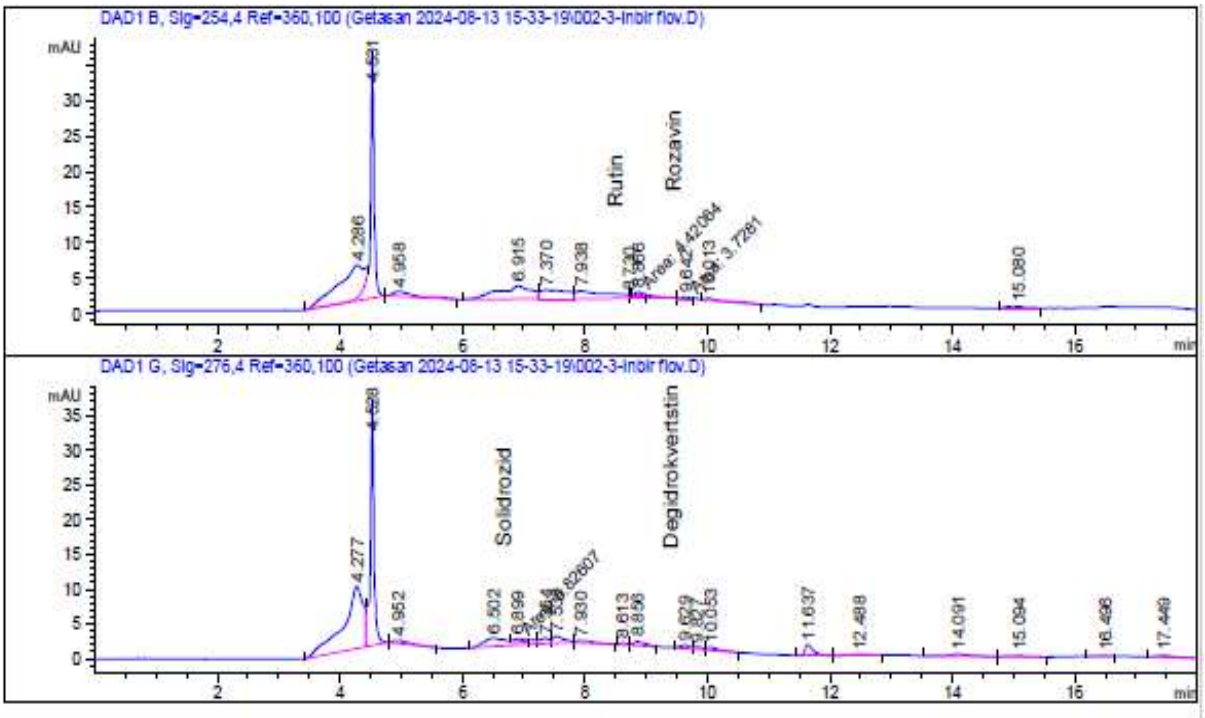


Fig. 3. HPLC-DAD chromatogram of the identified flavonoids in the *Z. officinale*

3.8. Elemental Composition Analysis: The mineral and trace element profile of the *Z. officinale* extract was determined using ICP-MS. The elemental content, including macro and trace elements, is shown in Table-6.

Table 6. Comprehensive Elemental Composition of *Z. officinale* Dry Extract (All values are expressed as mean ± SD, mg/kg, n=3)

No.	Element	Concentration (mg/kg)
1	Potassium (K)	120,000 ± 3,600
2	Magnesium (Mg)	120,000 ± 3,600
3	Phosphorus (P)	21,000 ± 630

No.	Element	Concentration (mg/kg)
4	Calcium (Ca)	1,800 ± 54
5	Manganese (Mn)	670 ± 20
6	Iron (Fe)	480 ± 14
7	Aluminium (Al)	230 ± 7
8	Zinc (Zn)	99.0 ± 3.0
9	Strontium (Sr)	13.0 ± 0.4
10	Copper (Cu)	12.0 ± 0.4
11	Lithium (Li)	7.50 ± 0.23
12	Vanadium (V)	3.00 ± 0.09
13	Rubidium (Rb)	2.30 ± 0.07
14	Lead (Pb)	2.30 ± 0.07
15	Nickel (Ni)	2.00 ± 0.06
16	Arsenic (As)	1.50 ± 0.05
17	Cobalt (Co)	1.10 ± 0.03
18	Chromium (Cr)	1.00 ± 0.03
19	Barium (Ba)	7.20 ± 0.22
20	Silver (Ag)	0.24 ± 0.01
21	Cadmium (Cd)	0.29 ± 0.01
22	Caesium (Cs)	0.15 ± 0.00
23	Uranium (U)	0.14 ± 0.00
24	Gallium (Ga)	0.11 ± 0.00
25	Bismuth (Bi)	0.09 ± 0.00
26	Thallium (Tl)	0.05 ± 0.00
27	Beryllium (Be)	0.02 ± 0.00
28	Mercury (Hg)	0.005 ± 0.00
29	Indium (In)	0.004 ± 0.00
30	Selenium (Se)	0.50 ± 0.02

The elemental analysis revealed a high concentration of essential macro-minerals, particularly potassium, magnesium and phosphorus, which significantly contribute to the nutritional profile of the *Z. officinale* extract. Trace elements, including manganese, iron and zinc, were detected within levels consistent with those reported for high-quality botanical extracts. Furthermore, heavy metals such as lead, arsenic and mercury were found to be within permissible safety limits, ensuring the extract's suitability for pharmaceutical and nutraceutical formulations.

CONCLUSION:

This study established a systematic approach for optimizing the extraction and chemical characterization of *Zingiber officinale* rhizome dry extract. Among the evaluated methods, ultrasonic-assisted extraction (UAE) achieved higher recovery of targeted secondary metabolites and nutrients in a shorter time than conventional maceration and percolation, which support its potential for efficient and scalable processing. HPLC-DAD analysis quantified the identified amino acids and flavonoids, with rosavin and dihydroquercetin among the major detected flavonoid constituents. ICP-MS analysis characterized the mineral profile, identifying K, Mg, Na, Fe, Zn and Mn as major essential elements. The concentrations of investigated potentially toxic elements remained within specified safety limits under the evaluated conditions. Combined chromatographic and elemental analyses provide a chemical basis for quality assessment and standardization of the extract. The standardized extract may serve as a starting material for nutraceutical development. Its incorporation into a polyherbal formulation containing *Physalis alkekengi* and *Armoracia rusticana* require further investigation of formulation stability, biological activity, safety and efficacy. These findings support further development of standardized ginger extracts for nutraceutical applications and may contribute to locally manufactured botanical products, potentially reducing dependence on imported formulations.

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