

Overview of chemical composition, pretreatment and extraction methods of bioactive compounds from *Polygonum multiflorum* root



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Abstract

Polygonum multiflorum Thunb. is a valuable traditional medicinal herb, widely used in Asian countries for treatment and health care. In recent years, this species has attracted research attention not only because of its bioactive compounds such as polyphenols, anthraquinones, and stilbene, but also because of its potential applications in functional foods and pharmaceuticals. This review systematically presents information on the chemical composition, secondary compounds, pretreatment methods, and extraction techniques from *P. multiflorum* roots. In particular, the article analyzes the advantages and disadvantages of traditional and modern extraction methods, the impact of each process on the recovery efficiency, and quality of active compounds. In addition, current limitations and future research directions are also discussed, aiming to provide a scientific basis for optimizing the processing procedure and effectively exploiting the value of *P. multiflorum* in the food and pharmaceutical industries.

Key words: biological activity, extract, functional food, *Polygonum multiflorum*, pretreatment.

Abbreviations: AC, antioxidant capacity; DGAT1, diacylglycerol O-acyltransferase 1; DW, dry weight; EAE, enzyme-assisted extraction; HDL-C, high-density lipoprotein cholesterol; HMG-CoA reductase, 3-hydroxy-3-methylglutaryl-CoA reductase; LDL-C, low-density lipoprotein cholesterol; MAE, microwave-assisted extraction; TPC, total polyphenol content; SEM, scanning electron microscopy; THSG, tetrahydroxystilbene-2-O-glucoside; TSG, 2,3,5,4'-tetrahydroxystilbene-2- β -glucoside; UAE, ultrasound-assisted extraction.

Introduction

Polygonum multiflorum Thunb. is an ancient medicinal herb in traditional Asian medicine, especially in China, Vietnam, Japan and Korea (Bounda, Feng 2015). *P. multiflorum* root is commonly used for its noted effects such as nourishing the liver and kidneys, anti-aging, enhancing vitality and improving blood circulation. (Ho et al. 2017). Over the past few decades, modern studies have provided scientific evidence for many biological activities of this species, including antioxidant, anti-inflammatory, neuroprotective, cardioprotective, and immunomodulatory activities (Zhang et al. 2018). However, despite these promising pharmacological properties, the practical use of *P. multiflorum* has been restricted by its well-documented hepatotoxicity. Recent studies have focused on processing and extraction strategies to remove or reduce hepatotoxic compounds, such as the work of Pang et al. (2024), which emphasized that toxicity control is essential for the safe application of this plant.

In terms of nutritional and chemical composition, *P.*

multiflorum root is rich in phenolic compounds (gallic acid, catechin, resveratrol, emodin), anthraquinone, flavonoid, stilbenoid, along with some amino acids, minerals and polysaccharides (Lee, Lee 2015). These compounds not only contribute to the medicinal value but also open up potential applications in the functional food and health beverage industry. Stilbene glycosides, anthraquinones, phenolics, phospholipids and carbohydrate compounds make up the main components of *P. multiflorum* (Saewan, Jimtaisong 2015).

Pretreatment of raw materials plays an important role in the quality of the extract and the final product. Steps such as cleaning, slicing, drying, grinding and storage can directly affect the stability and content of active compounds (Yang et al. 2021). In addition, traditional processing methods such as processing with black beans, steaming or soaking in alcohol can also change the chemical composition, thereby affecting the biological effects of *P. multiflorum* (Qian et al. 2024).

To maximize the value of *P. multiflorum* roots in food and pharmaceutical applications, selecting the appropriate

extraction method is crucial. Traditional techniques such as leaching or reflux are simple and easy to apply, but they are time-consuming, require a lot of solvents and often give low polyphenol recovery efficiency. In contrast, modern methods such as enzyme-assisted extraction (EAE), ultrasound-assisted extraction (UAE), or microwave-assisted extraction (MAE) help break down cell structures more effectively, shorten processing time, and improve extraction efficiency. In particular, when MAE is optimized for factors such as power, time, solvent ratio or pH, it can enhance polyphenol recovery while limiting thermal degradation of sensitive biological compounds (Orio et al. 2012).

Although each method has been studied individually, comparative evaluations across multiple techniques on the same raw material and under homogeneous analytical conditions remain limited. Therefore, this paper aims to provide a problem-oriented analytical review of studies on the pretreatment, chemical composition, and potential applications of *P. multiflorum*, with a particular focus on five extraction methods (EAE, MAE, UAE, reflux, maceration) (Wang et al. 2017). The analysis highlights differences in extraction efficiency for key bioactive compounds such as polyphenols, saponins, anthraquinones, and stilbenes, and further discusses the advantages, disadvantages, feasibility, and application potential of each method. By addressing both phytochemical value and safety concerns, this review provides a scientific basis for the selection and development of effective production processes for *P. multiflorum* root.

Chemical composition and biological activities of *P. multiflorum* root

Primary chemical composition

Table 1 shows that the chemical composition of *P. multiflorum* roots varies considerably, especially in respect to moisture, protein, and ash content. The water content is very high (78.61 – 96.82%), reflecting the characteristics of fresh roots and explaining why the material is susceptible to deterioration if not handled or stored promptly (Quoc, Muoi 2015b; Oh et al. 2018). Compared with other species of the same genus, *P. multiflorum* roots contain more water but have lower mineral content (ash, about 1.03 – 5.28%) than *Polygonum equisetiforme* (ash content of about 10.5%) and *Polygonum plebeium*. This may be due to differences

in tissue structure, growth environment and physiological functions of each species in the process of dry matter accumulation.

The protein content of *P. multiflorum* ranges from 2.13% (Vietnam) to 7.36% (Korea) (Quoc, Muoi 2015b; Choi et al. 2016). The differences between various studies may be due to soil conditions, climate, cultivars, harvest age, and analytical methods. In the context of food applications, this average protein value is not sufficient to consider *P. multiflorum* roots as a major source of protein, but it is still significant in terms of nutritional supplementation when processed into functional foods or medicinal materials.

The lipid content in *P. multiflorum* is relatively low (0.35 – 1.84%), consistent with the characteristics of roots, helping the raw material to be less susceptible to fat oxidation and prolonging the storage time after processing (Choi et al. 2016). However, the low lipid content also means that the energy value mainly comes from carbohydrates (such as starch and soluble sugar). In fact, the research of Quoc, Muoi (2015b) in Vietnam showed that starch content was about 8.88% and total carbohydrates 12.20%, indicating that the material can provide moderate energy, but is not a large source of starch compared to food crops such as potatoes with a starch content of 15 to 20% (Koch et al. 2018).

Compared with other *Polygonum* species, *P. plebeium* and *P. equisetiforme* stand out with higher protein and ash contents, which may open up the direction of hybridization or biochemical comparison to investigate the genetic and ecological factors affecting chemical composition (Batool et al. 2023; Auda 2025). In contrast, *P. multiflorum* is more interesting in respect to secondary compounds (polyphenols, anthraquinones, stilbene) than in its primary chemical composition. This difference determines the direction of use: species rich in minerals (calcium, magnesium, potassium, manganese, etc.) and nitrogen can be directed towards chemical supplements, while *P. multiflorum* is more suitable for the development of medicinal herbs and antioxidant products (Batool et al. 2021).

Overall, assessing the chemical composition of *P. multiflorum* root not only helps to understand the value of the raw material, but also supports the selection of appropriate processing and preservation methods. Due to its high water content, drying, steaming or heat

Table 1. Chemical composition of *Polygonum multiflorum* root and other *Polygonum* species

Species	Area	Part	Moisture	Protein (%)	Lipid (%)	Starch (%)	Ash (%)	Reference
<i>P. multiflorum</i>	Cheonan, Korea	Root	87.8 – 88.5	4.25 – 7.36	0.35 – 1.84	–	3.45 – 4.31	Choi et al. 2016
<i>P. multiflorum</i>	Vietnam	Root	78.61	2.13	0.42	8.88	1.03	Quoc, Muoi 2015b
<i>P. multiflorum</i>	Jeollanam-do, Korea	Root	91.75 – 96.82	4.29 – 7.13	0.57–1.11	–	4.62 – 5.28	Oh et al. 2018
<i>P. equisetiforme</i>	Israel	Leaves	8.8	7.1	1.2	–	10.5	Auda 2025
<i>P. plebeium</i>	Pakistan	Stem	6.46	8.75	6.79	–	–	Batool et al. 2023

treatment technologies – if well controlled – can help to preserve basic nutrients while facilitating the extraction of bioactive compounds (Rad et al. 2025). However, the large variability between data sources suggests that further direct comparative studies under the same conditions are needed to draw accurate conclusions and optimize the value of this raw material.

Secondary metabolites and biological activities

P. multiflorum is rich in secondary compounds, the most important of which are stilbene glycosides and anthraquinones (Table 2). Among them, 2,3,5,4'-tetrahydroxystilbene-2-O-glucoside (THSG) and anthraquinones are often chosen as indicator compounds to evaluate quality of this plant, with minimum contents of 1.0 and 0.10%, respectively (Lin et al. 2015). In addition to THSG, many other stilbenes have also been detected, such as tetrahydroxystilbene-O-(malonyl)-hex, tetrahydroxystilbene-O-deoxyhex and tetrahydroxystilbene-O-(caffeoyl)-hex; however, their structures have not been fully determined (Qiu et al. 2013).

The anthraquinone group is dominant with emodin-type derivatives, typically emodin, aloe-emodin, chrysophanol, etc. (Sun et al. 2013). These compounds play an important role in the biological activity of the medicinal plant, but are also related to the reported hepatotoxicity.

In addition, *P. multiflorum* also contains many flavonols

with strong antioxidant and free radical scavenging activities (Li et al. 2012). The flavonols identified include tricetin, rutin, luteolin, quercetin, etc. (Xu et al. 2006). Phospholipids are also present in significant amounts, which may contribute to the blood-enriching effects of the medicinal herb. Some representatives include phosphatidylethanolamine, copaeine, eicosane, hexanoic acid, etc. (Chen et al. 2001).

Besides, many other polyphenols such as 3-O-galloyl-(-)-catechin, 3-O-galloyl-(-)-epicatechin, 3-O-galloyl-procyanidin B2 and 3,3'-di-O-galloyl-procyanidin B2 are also present (Nonaka et al. 1982). Nitrogen-containing compounds have also been isolated, including *N-trans*-feruloyltyramine, *N-trans*-feruloyl-3-methyl-dopamine (Li et al. 1993) and indole-3-(L-amino-hydroxypropionic acid) methyl ester (Yang et al. 1998). In addition, two coumarin glucosides were also identified, 7-hydroxy-4-methylcoumarin-5-O- β -D-glucopyranoside and 7-hydroxy-3,4-dimethylcoumarin-5-O- β -D-glucopyranoside (Yu et al. 2008). Other compounds were also detected, including *n*-butyl- β -D-fructopyranoside (Zhang et al. 2006) and 1,3-dihydroxy-6,7-dimethylxanthone-1-O- β -D-glucopyranoside (Zhou et al. 1994).

In general, the diversity of secondary compounds has created a rich pharmacological basis for *P. multiflorum*, while also placing high demands on quality control and safety when applied in practice.

Table 2. Typical secondary metabolites identified in *Polygonum multiflorum* roots

Compound class	Representative compounds	Biological role/function	Reference
Stilbene glycosides	2,3,5,4'-Tetrahydroxystilbene-2-O-glucoside; tetrahydroxystilbene-O-(malonyl)-hex; tetrahydroxystilbene-O-deoxyhex; tetrahydroxystilbene-O-(caffeoyl)-hex	Antioxidant, neuroprotective; marker compounds for quality evaluation	Qiu et al. 2013; Lin et al. 2015
Anthraquinones	Emodin, aloe-emodin, chrysophanol, physcion	Anti-inflammatory, laxative; associated with hepatotoxicity	Sun et al. 2013
Flavonols	Tricetin, rutin, luteolin, quercetin, kaempferol, isoorientin, apigenin	Antioxidant, free radical scavenging	Xu et al. 2006; Li et al. 2012
Phospholipids	Phosphatidylethanolamine, copaeine, eicosane, hexanoic acid	Related to tonic and hematopoietic effects	Chen et al. 2001
Other polyphenols	3-O-galloyl-(-)-catechin, 3-O-galloyl-(-)-epicatechin, 3-O-galloyl-procyanidin B2, 3,3'-di-O-galloyl-procyanidin B2	Antioxidant, cytoprotective	Nonaka et al. 1982
Nitrogen-containing compounds	<i>N-trans</i> -feruloyltyramine, <i>N-trans</i> -feruloyl-3-methyl-dopamine, indole-3-(L-amino-hydroxypropionic acid) methyl ester	Potential biological activities	Li et al. 1993; Yang et al. 1998
Coumarin glycosides	7-hydroxy-4-methylcoumarin-5-O- β -D-glucopyranoside, 7-hydroxy-3,4-dimethylcoumarin-5-O- β -D-glucopyranoside	Antioxidant, circulatory support	Yu et al. 2008
Xanthone glycoside	1,3-dihydroxy-6,7-dimethylxanthone-1-O- β -D-glucopyranoside	Potential biological activity	Zhou et al. 1994
Sugars and other glycosides	<i>n</i> -butyl- β -D-fructopyranoside	Possible role in energy metabolism	Zhang et al. 2006

Pretreatment of *P. multiflorum* root

Pretreatment during phenolic extraction plays an important role in preserving or improving the chemical composition of *P. multiflorum* roots before extraction (Table 3). The raw material is usually harvested at the mature stage, 15 to 25 cm long, 5 to 10 cm in diameter, and free of pests and diseases. After washing under running water and brushing the surface to remove impurities, the roots can be processed in many different ways. The method of slicing thinly (2 to 3 mm) and drying at 60 °C to < 12% moisture before grinding (< 0.5 mm) and vacuum packaging helps to preserve long-term and limit the growth of microorganisms. Compared with fresh samples, dried samples of *P. multiflorum* showed a 49 to 62% decrease in total polyphenol content (TPC) and a 71 – 78% decrease in antioxidant capacity (AC) (Quoc, Muoi 2015b). This deterioration is mainly due to the impact of heat during the drying process, which decomposes or changes the structure of phenolic compounds, combined with oxidation during processing and storage, leading to significant loss of bioactive substances.

In the processing of medicinal materials, the nine cycles of steaming and sun-drying method is applied to reduce toxicity and improve taste. This process includes slicing (3 mm), steaming for 4 h, holding for 10 h, and drying at 40 °C, repeated nine times. Qian et al. (2020) showed that polysaccharide content decreased from 162.18 mg g⁻¹ (fresh sample) to 97.72 mg g⁻¹ after processing, while fructose and glucose increased sharply in the early cycles and then decreased slightly in the final stage. Saponin content increased steadily from 27.06 to 46.50 mg g⁻¹, while flavonoid and polyphenol contents fluctuated with

the thermal cycle, reaching their maximum at 22.00 and 8.35 mg g⁻¹, respectively. This change reflects the dual effect of prolonged temperature and time, which both promotes hydrolysis and chemical transformation, and causes partial loss of thermosensitive compounds.

In addition, some milder pretreatment methods such as shade drying, low temperature drying (< 50 °C) or mechanical grinding are applied to limit active ingredient loss, especially in small scale or manual production (Thang et al. 2017). However, these methods often result in shorter storage times and are susceptible to microbial contamination if the final moisture content is not well controlled.

In general, the choice of pretreatment method depends on the final objective: if the priority is to retain the active compounds to the maximum, processing at moderate temperatures and short times is necessary; if the goal is to reduce toxicity and improve the sensory appearance, multi-cycle thermal processes such as nine cycles of steaming and sun-drying remain the traditional choice, although they come with the risk of some loss of chemical value.

Extraction methods

Reflux extraction (maceration)

Reflux is a traditional technique that uses a heated solvent to continuously circulate through the material to dissolve the desired compound. Although simple and low cost, this method requires a long time and often does not completely disrupt the cell structure, resulting in limited extraction efficiency compared to modern techniques (Olalere, Gan 2021).

Table 3. Comparison of pretreatment methods of *Polygonum multiflorum* roots. AC, antioxidant capacity; TPC, total polyphenol content

Method	Processing conditions	Impact on chemical composition	Advantage	Application objectives	Reference
Dry at 60 °C after slicing 2 – 3 mm, grind finely, vacuum pack	Dry at 60 °C to moisture < 12%, grind < 0.5 mm	TPC decreased by 49 – 62%, AC decreased by 71 – 78% compared to fresh samples	Long-term preservation, limited microorganisms, easy to transport	Prepare raw materials for extraction or powder production	Quoc, Muoi 2015a
Sun-drying, mechanical grinding, sieving	Dry in cool place, avoid direct sunlight	Little effect on heat sensitive compounds; slight oxidation risk	Easy to implement, low cost, suitable for small scale	Handcrafted, locally processed	Thang et al. 2017
Mechanical grinding from dried roots	Chop, and grind finely before extracting.	No change in composition, only increase in surface area	Fast, no chemical change	Increased extraction efficiency by increasing contact area	Choi et al. 2016
Nine cycles of steaming and sun-drying	Cut into 3 mm slices, steam for 4 h, keep for 10 h, dry/expose at 40 °C, repeat 9 times	Polysaccharides decreased from 162.18 to 97.72 mg g ⁻¹ ; saponins increased from 27.06 to 46.50 mg g ⁻¹ . Flavonoid, polyphenol fluctuated, reaching maximum between cycle	Reduce toxicity, improve taste, change beneficial chemical composition	Processing medicinal herbs according to traditional medicine	Qian et al. 2024
Low heat drying (< 50°C)	Dry to moisture < 12%	Reduced polyphenol loss compared to 60 °C, preserving flavor	Maintain high quality, limit chemical changes	Processing high-quality products, keeping natural flavors	Wang et al. 2017

In a trial using *P. multiflorum*, root powder was extracted with 60% acetone solvent at a raw material/solvent ratio of 1:40 (w/v) for 90 min. The results obtained TPC of 38.60 ± 0.56 mg GAE g⁻¹ DW (gallic acid equivalents per gram dry weight) and AC of 298.15 ± 2.99 μ mol TE g⁻¹ DW (Trolox equivalents per gram dry weight), in which some compounds such as catechin (1.98 mg g⁻¹), gallic acid (0.58 mg g⁻¹), and resveratrol (0.023 mg g⁻¹) were detected. Scanning electron microscopy (SEM) observation showed that the cells were less deformed and the tissue structure was intact, indicating that the process was mainly based on passive dissolution instead of mechanical disruption (Quoc 2020).

In addition to phenolic compounds, a characteristic and highly abundant compound of *P. multiflorum*, 2,3,5,4'-tetrahydroxystilbene-2-O- β -d-glucoside (TSG), has been obtained by aqueous extraction (Zhang et al. 2017). The optimal procedure involves soaking the raw material in water at a ratio of 1:12 for 30 min, followed by boiling for 70 min with multiple extraction cycles.

The advantages of the reflux method are simple equipment, low investment costs, and ease of operation. However, the disadvantages are long extraction times, high energy and solvent consumption, and easy thermal degradation of sensitive compounds. This method is still suitable for small-scale production or when facilities are limited (Sun et al. 2021).

α -Amylase-assisted extraction

The method using the enzyme α -amylase takes advantage of the ability to hydrolyze starch into smaller sugar molecules, helping to break down the carbohydrate storage structure in the root tissue, thereby releasing polyphenols and other active compounds. This process is especially useful for materials with high starch content, which can hinder the diffusion of compounds into the solvent (Wang et al. 2010).

In the study, *P. multiflorum* root powder was treated with α -amylase at a concentration of 0.3% (v/w), material/solvent ratio of 1:10 (w/v), pH 6, temperature 80 °C for 2.5 h (Quốc, Mười 2015a). The results achieved TPC of 23.55 ± 1.84 mg GAE g⁻¹ DW and AC of 131.62 ± 3.34 μ mol TE g⁻¹ DW. High performance liquid chromatography analysis showed the presence of resveratrol (0.017 mg g⁻¹), gallic acid (0.26 mg g⁻¹), and catechin (1.57 mg g⁻¹). After treatment, the material completely changed in morphology, proving the effectiveness of breaking down the structure of α -amylase enzyme (Quốc, Mười 2015a).

The advantage of this method is to increase the release of polyphenols bound to the starch matrix, reduce the viscosity of the extract, and facilitate the diffusion of compounds. However, the disadvantage is that it requires high temperatures to activate the enzyme, which can cause the decomposition of some heat-sensitive compounds. In addition, the extraction time is quite long and the cost of enzymes is still a barrier to expanding to production scale (Dhital et al. 2017).

Pectinase-assisted extraction

Pectinase-assisted extraction is an approach that is receiving much attention in the exploitation of bioactive compounds from medicinal herbs, especially for pectin-rich root species such as *P. multiflorum* (Choi et al. 2015). Pectinase is a group of enzymes that catalyze the hydrolysis of pectin polysaccharide chains and glycosidic bonds in plant cell walls. Pectin acts as an “adhesive” between cells, binding with cellulose, hemicellulose and protein into a solid system, protecting the secondary compounds inside (Haile, Ayele 2022). When pectin is degraded, the tissue structure is broken and the distance between microcapillaries in the cell wall increases, creating favorable conditions for the solvent to penetrate deeply and carry away soluble compounds, thereby improving the efficiency of collecting polyphenols, flavonoids, and stilbene glycoside compounds characteristic of *P. multiflorum* (Larsen et al. 2021).

In the study of Quoc, Muoi (2017), the pectinase extraction process was optimized with a raw material/solvent ratio of 1:11 (w/v), enzyme concentration of 0.2% (v/w), pH 4.5, temperature of 50 °C, and time of 80 min. This is a balance between maximum pectinase activity and stability of phenolic compounds, helping to limit thermal decomposition. The results showed that the TPC reached 44.36 mg GAE g⁻¹ DW and the AC (DPPH) reached 80.43 μ mol TE g⁻¹ DW. HPLC analysis identified a number of major phenolic compounds such as gallic acid (3.65 mg g⁻¹) and catechin (1.56 mg g⁻¹), two active ingredients with strong AC and anti-inflammatory and neuroprotective potential. SEM observation showed that the tissue surface was completely broken, the cells collapsed, and many voids appeared, proving the combined mechanical and chemical effects of pectinase.

The outstanding advantages of this method are high efficiency under moderate temperature conditions, environmental friendliness thanks to the use of water or dilute organic solvents, and limited energy use compared to traditional heating methods. Avoiding high temperatures also helps preserve the structure and biological activity of easily degradable compounds, especially heat-sensitive stilbene glycosides (Shimoda et al. 2015). In addition, this method can be combined with other supporting techniques such as ultrasound (UAE) or microwave (MAE) to shorten the time and improve the efficiency of compound release.

With such potential, pectinase-assisted extraction is not only suitable for laboratory-scale medicinal research but also has the potential to expand its application in the production of functional foods, herbal beverages, or pharmaceutical ingredients. However, the cost of enzymes and the requirement for strict pH-temperature control are challenges that need to be addressed when transferring to an industrial scale. In the context of the trend of developing natural, safe, and environmentally friendly products, this method promises to continue to receive attention and improvement in the future.

Ultrasound-assisted extraction (UAE)

Ultrasound-assisted extraction (UAE) is a method of extracting active compounds based on the cavitation effect – the formation, growth, and collapse of gas bubbles in a liquid medium under the influence of high-frequency ultrasonic waves (Panda, Manickam 2019). When the air bubbles collapse, they create strong microcurrents and shockwaves that are strong enough to break the cell walls, increase the contact area between the solvent and the raw material, and promote the diffusion of dissolved compounds. With the advantage of direct and uniform energy transmission, UAE allows for improved extraction efficiency in a short time and at low temperatures, limiting the decomposition of heat-sensitive compounds (Sanjaya et al. 2022).

In a study on *P. multiflorum*, root powder was mixed with 60% acetone solvent at a material/solvent ratio of 1:30 (w/v) and treated in an ultrasonic bath with a power of 550 W at 60 °C for 15 min (Quoc, Muoi 2016). This condition resulted in a TPC of 43.28 ± 0.54 mg GAE g⁻¹ DW and an AC (DPPH) of 343.88 ± 3.06 µmol TE g⁻¹ DW. The phenolic compounds detected included resveratrol (0.028 mg g⁻¹), gallic acid (0.35 mg g⁻¹), and catechin (1.83 mg g⁻¹). SEM images showed severe cell destruction, torn cell walls, and obvious collapsed tissue structures, demonstrating the mechanical effect of ultrasound during the extraction process (Quoc, Muoi 2016).

In addition to phenolic compounds, some authors have focused on the stilbene glycosides group, a characteristic component of *P. multiflorum*. For example, Denget al. (2025) applied an ethanol-assisted ultrasound extraction process. The optimal process included 60% ethanol concentration, 25 g mL⁻¹ solid-liquid ratio, 65 °C temperature, 1.5 h, and 800 W, resulting in a stilbene glycosides recovery efficiency of 7.19%. Notably, the non-heated ultrasound extraction could limit the degradation of stilbene glycosides, showing that this method not only has technical advantages but also has potential for industrial application. The outstanding advantages of UAE are short extraction time, reduced solvent consumption, preservation of active ingredients and easy application on a semi-industrial scale. However, the main limitations are the high investment cost of high-power ultrasonic equipment and high power consumption. In addition, if the power or processing time is too high, the local temperature increase may denature or decompose some sensitive compounds (Tiwari 2015). UAE is currently considered a potential method for the extraction of bioactive compounds from *P. multiflorum*, especially when fast processing time and high quality of active ingredients are required.

Microwave-assisted extraction (MAE)

Microwave-assisted extraction (MAE) is based on the principle that microwave radiation directly affects polar molecules such as water and ethanol, causing them to

vibrate strongly and generate internal heat. This volume heating increases the pressure inside the cell, disrupts the cell membrane and releases the active compound into the solvent (Ridlo et al. 2020). Unlike conventional heating, MAE heats the entire sample evenly, significantly shortening extraction time and reducing the amount of solvent required (Mandal et al. 2007).

In a study with *P. multiflorum*, root powder was extracted using an industrial microwave at 127 W, 60% acetone solvent, material/solvent ratio 1:40 (w/v) for 5 min (Quoc, Muoi 2016). The results showed TPC of 44.3 ± 0.13 mg GAE g⁻¹ DW and AC of 341.26 ± 1.54 µmol TE g⁻¹ DW. The phenolic compounds detected included gallic acid (0.78 mg g⁻¹), catechin (5.58 mg g⁻¹), and resveratrol (0.12 mg g⁻¹). SEM images showed strong cell deformation, cracked cell walls, and gelatinized starch inside – evidence of a rapid heating effect and high intracellular pressure of MAE (Quoc, Muoi 2016).

In addition to the ability to effectively recover phenolic compounds, MAE is also applied to extract TSG and stilbene derivatives, typically as in the study of Qian et al. (2020) using ethanol solvent combined with electromagnetic waves to increase extraction speed. The time is only 10 min, saving 50% of solvent compared to the traditional reflux method. TSG and stilbene derivatives were determined by UV-Vis spectroscopy and HPLC-DAD.

The advantages of MAE are extremely short time, saving solvent and achieving high efficiency. However, the biggest disadvantage is that it is difficult to control the temperature if the microwave power exceeds the optimal threshold, leading to the decomposition of phenolic compounds and reducing product quality (Serdar et al. 2017). In addition, specialized MAE equipment is expensive and must be operated under microwave radiation safety conditions. With high efficiency and the ability to preserve active compounds in a short time, MAE is the right choice when rapid production with maximum efficiency is required (Zhao et al. 2018).

Extraction by a Soxhlet system

The Soxhlet method is a hot solvent extraction technique that involves repeated cycles, allowing fresh solvent to continuously contact the material, ensuring complete extraction of the desired compound. The Soxhlet method is often used for total extraction, including phenolic compounds, alkaloids and lipids (Ramluckan et al. 2014).

In the application on root material, about 5 to 50 g of powder was extracted with organic solvents of increasing polarity (n-hexane, ethyl acetate, methanol) or mixed solvents (ethanol 80%, butanol) for 6 to 12 h. After extraction, the solvent was removed by rotary evaporation, and the extracted powder was freeze-dried for storage. TPC yield can range from several hundred to more than 1200 mg GAE g⁻¹ depending on solvent and material, and AC (DPPH) reaches over 70 – 89% (Choi et al. 2016).

The advantages of the Soxhlet method are the ability to exhaust and reuse the circulating solvent, suitable for comprehensive chemical composition studies. However, the major disadvantages are the long extraction time, high solvent and energy consumption, and the high risk of decomposition of thermosensitive compounds (López-Bascón, Castro 2020). Therefore, the Soxhlet method is currently used mainly in analytical research rather than large-scale production.

Summary of extraction methods

The comparison of results showed that different extraction methods yielded significant variations in TPC and AC of *P. multiflorum* root extracts (Table 4). Among the state-of-the-art techniques, MAE, and UAE achieved similar TPC (44.36 and 43.28 mg GAE g⁻¹ DW) with very high AC (> 340 µmol TE g⁻¹ DW), while having short processing times (5 to 15 min), reflecting the effective tissue disruption by volumetric heating (MAE) or cavitation (UAE) (Kumar et

al. 2021; Pereira et al. 2023). Pectinase-assisted extraction also achieved high TPC (44.36 mg GAE g⁻¹ DW) but lower AC (~80 µmol TE g⁻¹ DW), indicating that this enzymatic method effectively released polyphenols but the release rate of compounds with strong antioxidant capacity was lower than that of UAE or MAE (Quoc, Muoi 2017; Quoc, Muoi 2018).

In contrast, leaching has the advantage of cost and simple equipment, but TPC and AC are significantly lower, mainly due to the long extraction time (90 min), but does not create mechanical breaking force, leading to limited compound recovery (Quoc 2020). α-Amylase-assisted extraction gave the lowest TPC and AC, possibly due to the high temperature (80 °C) and long time (2.5 h) causing partial degradation of thermosensitive phenolic compounds, although this method was valuable in recovering starch-bound polyphenols (Quoc, Muoi 2015a). Soxhlet extraction gives very high TPC (up to 1454.5 mg GAE g⁻¹), but involves a long extraction time and high

Table 4. Comparison of extraction methods for phenolic compounds from *Polygonum multiflorum* roots. TPC, total polyphenol content; GAE, gallic acid equivalents; AC, antioxidant capacity; TE, trolox equivalents; DW, dry weight

Method	Optimal conditions	Result	Main compound	Advantage	Reference
Pectinase-assisted extraction	Raw material/solvent ratio 1:11 (w/v); enzyme concentration 0.2% (v/w); pH 4.5; 50 °C; 80 min	TPC 44.36 mg GAE g ⁻¹ DW; AC 80.43 µmol TE g ⁻¹ DW	Gallic acid (3.65 mg g ⁻¹), catechin (1.56 mg g ⁻¹)	High efficiency; effective cell wall disruption; environmentally friendly; moderate temperature	Quoc, Muoi 2017
Ultrasound-assisted extraction	Acetone 60%; ratio 1:30 (w/v); 60 °C; power 550 W; 15 min	TPC 43.28 ± 0.54 mg GAE g ⁻¹ DW; AC 343.88 ± 3.06 µmol TE g ⁻¹ DW	–	Short time; preserve active ingredient; reduce solvent	Quoc, Muoi 2018
Microwave-assisted extraction	Acetone 60%; ratio 1:40 (w/v); power 127 W; 5 min	TPC 44.3 ± 0.13 mg GAE g ⁻¹ DW; AC 341.26 ± 1.54 µmol TE g ⁻¹ DW	–	Extremely short time; solvent saving; high efficiency	Quoc, Muoi 2016
	–	–	2,3,5,4'-Tetrahydroxystilbene-2-O-β-d-glucoside and stilbene derivatives	Time only 10 min, save 50% solvent	Wang et al. 1994
Reflux extraction	Acetone 60%; ratio 1:40 (w/v); 90 min	TPC 38.60 ± 0.56 mg GAE g ⁻¹ DW; AC 298.15 ± 2.99 µmol TE g ⁻¹ DW	–	Simple equipment; low cost	Quoc, Muoi 2020
	Raw materials in water at a ratio of 1:12 for 30 min, then boiled for 70 min with multiple extraction cycles.	–	2,3,5,4'-Tetrahydroxystilbene-2-O-β-d-glucoside		Zhang et al. 2017
α-Amylase-assisted extraction	Ratio 1:10 (w/v); enzyme concentration 0.3% (w/v); pH 6; 80 °C; 2.5 h	TPC 23.55 ± 1.84 mg GAE g ⁻¹ DW; AC 131.62 ± 3.34 µmol TE g ⁻¹ DW	Resveratrol (0.017 mg g ⁻¹), gallic acid (0.26 mg g ⁻¹), catechin (1.57 mg g ⁻¹)	Release of starch-bound polyphenols; reduction of extract viscosity	Quoc, Muoi 2015a
Soxhlet extraction	5 – 50 g raw material; solvent <i>n</i> -hexane, EtOAc, MeOH or 80% ethanol; 6 – 12 h	TPC 1212.6 – 1454.5 mg GAE g ⁻¹ ; AC 73.1 – 89% (DPPH)	–	Extractive; suitable for studying total composition	Choi et al. 2016

temperature risk changing the compound structure, and consumes a lot of solvent and energy, thus making it less suitable for sensitive compounds (Choi et al. 2016).

In general, physically assisted (UAE, MAE) and enzymatic (pectinase) methods stand out for their high yield, short time or mild conditions, suitable for preserving bioactivity. Meanwhile, traditional methods (Reflux, Soxhlet) are still useful when total recovery of the component is required or for small-scale deployment, but need to be optimized to reduce thermal and solvent effects. The choice of method depends on the end product objective: if quality and bioactivity are the priority, UAE or pectinase are suitable options; if exhaustive extraction is the priority, The Soxhlet method can be considered for research scale operations.

Toxicity of *P. multiflorum* root

In recent years, the safety of using medicinal herbs in general and *P. multiflorum* in particular has received increasing attention (Hu, Caldach 2017). Several reports have noted toxicity, especially with prolonged use (Lin et al. 2015). Anthraquinones and their derivatives are considered to be the main contributors, with concentrations in the roots reaching up to 2.51% (Feng et al. 2017). Some derivatives such as rhein, emodin, aloe-emodin show high cytotoxicity, related to their ability to bind to DNA and glutathione (Panigrahi et al. 2018).

Tests on zebrafish embryos showed that anthraquinone, anthrone and naphthol were severely toxic, while stilbene glycoside was virtually non-toxic (Yang et al. 2018). Similar, Lv et al. (2015) confirmed on human liver cells LO2 that emodin-8-O- β -D-glucopyranoside, physcion-8-O- β -D-glucopyranoside, emodin and physcion were all significantly toxic, and emphasized that the toxicity depended on the extraction solvent: ethanol extract gave stronger toxicity than water.

However, there have been also toxicity studies in rodents that have not noted serious effects. Min et al. (2025) reported that the lethal dose of both powder and decoction exceeded 5000 mg kg⁻¹. Repeated toxicity studies over 13 weeks did not show major changes, except for gastric hyperplasia and renal tubular basophilia at 5000 mg kg⁻¹ day⁻¹.

This difference is largely due to the origin of the raw material, the state of processing, and the extraction method. Raw materials and organic solvent extractions (especially ethanol) generally give higher toxicity than processed medicinal materials or extracts with mild solvents. Wu et al. (2012) showed that processing reduced 2,3,4',5-tetrahydroxystilbene-2-O- β -D-glucoside by 55.8% and increased emodin by 34.0%, which may affect toxicity in both directions.

To reduce toxicity, some authors have studied selective separation techniques for anthraquinones. Yuan et al. (2020)

proposed to use ionic liquid combined with ultrasound, achieving an anthraquinone extraction efficiency of 97.2% and satisfactory solvent recovery. Similarly, Pang et al. (2024) developed deep eutectic solvents as a green and efficient strategy combined with ultrasound-assisted extraction, not only improving the extraction of beneficial phytochemicals but also effectively reducing hepatotoxic constituents, highlighting the importance of solvent design in enhancing both efficacy and safety.

The toxicity of *P. multiflorum* depends not only on the chemical composition but also on the origin of the raw materials and the extraction method. Raw materials and organic solvent extraction often give higher toxicity than processed medicinal materials or extraction with mild solvents. This difference may stem from changes in the content of anthraquinone and stilbene glycoside during processing, as well as the selective solubility of each extraction method. In addition, some works have noted that some compounds in the extract from *P. multiflorum* tubers have hepatoprotective effects (Huang et al. 2007). Therefore, safety assessment needs to be conducted comprehensively, combining many different criteria and test conditions before putting into practical application.

Applications of *P. multiflorum* root

In Vietnam, *P. multiflorum* has long been used in traditional medicine with many uses such as nutrition, treating impotence, reducing cholesterol and blood sugar, anti-aging, sedative, treating fever, rheumatism, headaches, tumours, and as an antibacterial (Ho 1999).

In China, the species has been used in traditional medicine for centuries as a liver and kidney tonic, longevity enhancer, and hair darkener. These health benefits have long been exploited through a variety of commercial products such as root powder, extracts, capsules, tinctures, shampoos, and body sprays (Ho et al. 2019).

Modern research has confirmed many of these traditional effects, showing that active compounds such as 2,3,5,4'-tetrahydroxystilbene-2- β -glucoside (TSG), emodin and polysaccharides in *P. multiflorum* have the ability to reduce total cholesterol, triglycerides, and LDL-C, while increasing HDL-C, inhibit lipid synthesis enzymes such as HMG-CoA reductase and DGAT1, stimulate lipolysis, improve insulin resistance, reduce blood sugar, and limit liver fat accumulation (Lin et al. 2015; Jung et al. 2020). This evidence demonstrates that the pharmacological value of *P. multiflorum* is fully consistent with its traditional application, and at the same time provides a scientific basis for the development of preparations to support the treatment of metabolic disorders.

P. multiflorum has long been used as a spice or medicinal herb in daily meals to improve health. Common processing methods include making spice powder, cooking

with chicken, steaming with pork liver, making soup, making tea or soaking in wine (Li, Gao 2015). In Vietnam, *P. multiflorum* is also widely used in folk cuisine. This medicinal herb is often stewed with chicken, pigeon or pork to make nutritious soup. In addition, *P. multiflorum* is also used to soak medicinal wine, make herbal tea, cook health-preserving tea or grind into powder and mix into porridge, cereal powder, and medicinal cakes. These processing methods not only help reduce the natural toxicity of the root, but also bring a unique flavor and health value to the dish.

Potential and challenges of *P. multiflorum*

P. multiflorum is a precious medicinal source rich in bioactive compounds, with chemical value and diverse applications in cuisine and medicine. Hundreds of different compounds have been identified from this species, many of which have special structures and high biological potential (Wang et al. 2023). This is an important basis for research on the development of functional products, cosmetics, and pharmaceuticals of natural origin. Expanding the planting area and applying technological advances in extraction and isolation can increase economic value and improve the capacity to exploit this resource.

In Vietnam, the main source of *P. multiflorum* raw material is still from wild plants. In addition to the purpose of exploiting the roots as medicinal materials in traditional medicine, it is also grown as a trellis for shade in gardens or planted along fences to create landscapes and windbreaks, while helping to retain soil and prevent erosion in mountainous areas. This leads to inconsistencies in quality and chemical composition due to the influence of regional ecological conditions. In addition, the long growth and harvesting time (usually over 3 to 4 years) results in low yields, while exploitation conditions are difficult because the plant often grows in remote mountainous areas. The price is therefore relatively high (7 to 9 USD for kg), limiting the possibility of mass use.

Although many valuable compounds have been identified, the isolation of individual compounds from *P. multiflorum* remains a challenge due to its diverse and complex chemical structure, requiring modern refining technology and large investment costs. In addition, factors such as unstandardized harvesting, preservation and transportation techniques can also cause changes in active ingredients, reducing chemical value and biological effects. Therefore, comprehensive studies on varieties, cultivation, harvesting and post-harvest processing are needed to ensure stable quality and maximize the economic value of this plant species.

Conclusions

The synthesis of research results shows that *P. multiflorum* has a normal nutritional value but is notable for its high

content of active biological compounds, especially polyphenols and stilbene, which brings great potential for application in antioxidant and health protection products. The chemical composition and biological activity are strongly influenced by growth conditions, pretreatment methods and extraction techniques. Among them, modern methods such as MAE, UAE and pectinase enzyme extraction show advantages in terms of efficiency, time and ability to preserve heat-sensitive compounds, suitable for industrial-scale production. In contrast, traditional techniques such as reflux and soxhlet still have a certain role in basic research and exploitation of the entire chemical composition. The choice of the optimal process should be based on the final product target, while considering economic, environmental and raw material characteristics. Future research should focus on standardizing pretreatment procedures, evaluating the effects on major compound groups, and developing sustainable extraction technologies, in order to more effectively exploit the value of *P. multiflorum* in the functional food and pharmaceutical industries.

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