







## Influence of soil and climatic conditions on the chemical composition value of *Valeriana officinalis*

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### ABSTRACT

Chlorpyrifos (CPF) is an organophosphate pesticide widely used in agriculture. It poses significant health risks due to the leaves and roots of *Valeriana officinalis* harboring various chemical compounds, which are subject to alteration by environmental conditions and soil composition. The acidity of the soil plays a critical role in the absorption of essential nutrients, thereby modifying the concentrations of bioactive constituents such as valerenic acid. Furthermore, macro-nutrients, including nitrogen, phosphorus, and potassium, are vital for the plant's biosynthesis of these substances. Climatic factors, including temperature, humidity, and sunlight exposure, also affect the biochemical pathways of the plant, leading to variability in its chemical profile. Moreover, fluctuations in temperature can impact the levels of beneficial compounds, while water scarcity may induce the synthesis of stress-related compounds, which in turn affect the therapeutic properties of the plant. Therefore, enhancing both soil conditions and climatic factors could contribute to augmenting the medicinal efficacy of *V. officinalis*. This research investigates the influence of soil and climatic factors on the chemical composition of *V. officinalis*, a medicinal herb recognized for its therapeutic benefits. The leaves and roots of this species harbor a variety of chemical compounds that environmental conditions and soil characteristics can modify. The soil's acidity is particularly significant in nutrient uptake, which alters the concentrations of bioactive components, especially valerenic acid, a key element for the herb's medicinal effectiveness. Essential macronutrients such as nitrogen, phosphorus, and potassium are crucial for the production of these advantageous compounds, suggesting that effective soil management practices can improve the quality of the plant. Furthermore, climatic elements, including temperature, humidity, and sunlight exposure, play a critical role in the biochemical processes of *V. officinalis*, resulting in variations in its chemical makeup. Temperature variations can influence the concentration of beneficial compounds, while limited water availability may trigger the production of stress-related substances, thereby affecting the plant's therapeutic attributes. The results indicate that enhancing both soil quality and

climatic conditions could substantially improve the medicinal properties of plants. This study highlights the significance of environmental management in the cultivation of this important herb to maximize its therapeutic potential for consumers.

**Keywords:** Valeriana officials, Season, Temperature, Culture, Extraction

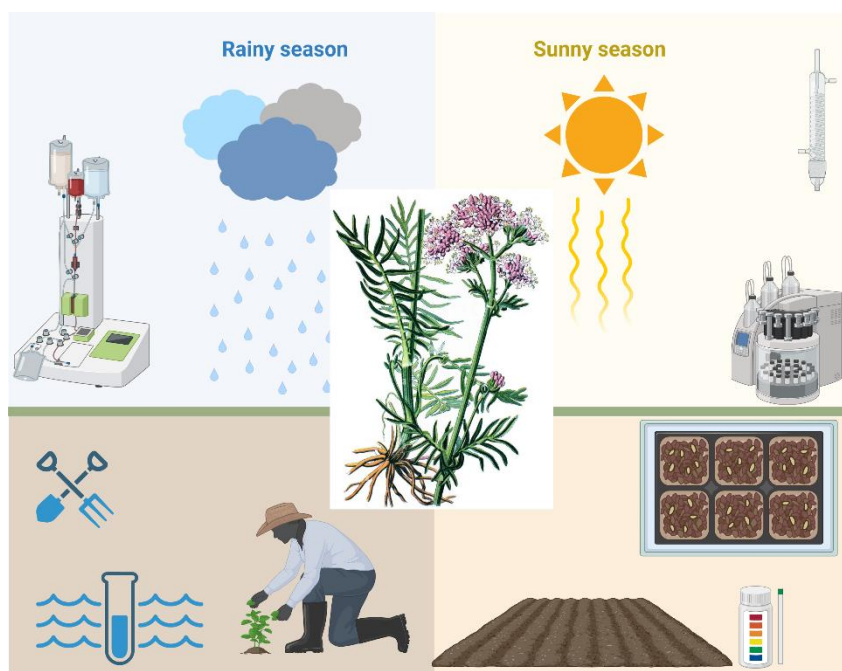
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## INTRODUCTION

Common valerian, *Valeriana officinalis* L. is a perennial herbaceous plant of medicinal significance that possesses a well-documented history of therapeutic efficacy. Its cultivation has expanded commercially across northern Europe, the Americas, and, more recently, Australia. This species ranks as Europe's fourth most commercially successful medicinal herb, boasting retail revenues of US\$ 200 million. The plant is characterized by robust subterranean rhizomes that generate numerous cylindrical roots, which are utilized for their sedative effects, addressing conditions such as nervous tension, insomnia, anxiety, and stress. Additionally, it has applications in managing hypertension and cardiac arrhythmias (Patočka & Jakl 2010; Haydarov *et al.* 2022). A critical parameter for assessing the quality of valerian raw material is the concentration of active constituents that confer health benefits to consumers. Although ongoing debate exists regarding the comparative efficacy of various compound classes, it is broadly acknowledged that valerenic acid (found within its essential oil) constitutes the most significant biologically active component, attracting considerable industrial interest in enhancing its concentration in valerian raw material. Essential oil is situated within the hypodermis in extensive thin-walled cells located at the periphery of the roots and rhizomes. Furthermore, valerian raw material is abundant in valepotriates, alkaloids, caffeic acid derivatives, flavonoids, lignans, and amino acids (Aparecida Gelfuso *et al.* 2014). Nonetheless, Wills & Shohet (2003) indicated a distinction in quality between rootlets and crowns (rhizomes). Nevertheless, numerous valerian producers need help meeting the pharmaceutical industry's stringent requirements, particularly concerning the elevated levels of active substances. As a result, research should be conducted not only to identify the most productive variety that simultaneously possesses an adequate concentration of active compounds but also to delineate optimal cultivation principles, including the identification of the most effective plantation establishment methods, which are crucial for ensuring high-quality raw materials. It is recognized as one of the largest producers of valerian, the species is typically propagated through seed sowing in seedbeds, followed by the transplantation of seedlings into the field (Wills & Shohet 2003). There is a conspicuous absence of literature detailing the establishment of valerian plantations utilizing direct seed sowing in the field or the production of seedlings in greenhouse settings employing polypropylene multi-cell trays. The latter methodology facilitates uniform plant distribution, enhanced light access, improved humidity conditions, and minimized risk of root damage during transplantation (Raj *et al.* 2023). Consequently, the yields from plants propagated using this technique are approximately 15% greater than those derived from seedlings cultivated in seedbeds. The objective of the present study was to identify the most appropriate variety of common valerian (distinguishing between thin- and thick-rooted types) and to assess the impact of the plantation establishment method on the development of subterranean structures (roots and rhizomes), yield outcomes, and the concentrations of essential oil and valerenic acid (in roots and rhizomes) as indicators of raw material quality (Wiśniewski *et al.* 2016). The chemical composition of *V. officinalis* is significantly influenced by soil and climatic conditions, which affect its growth and phytochemical profiles (Fig. 1). Variations in environmental factors lead to differences in the concentration of essential oils, phenolic compounds, and other bioactive constituents. The following sections elaborately describe these influences. The presence of macro- and microelements in the soil directly impacts the chemical composition of *V. officinalis*. Studies have shown that variations in soil nutrients can lead to differences in the content of essential oils and secondary metabolites. Different soil types, such as those found in mountainous versus pre-mountainous regions, can affect the plant's mechanical tissue development and overall quality (Letchamo *et al.* 2004). Climatic factors such as temperature and humidity influence the synthesis of secondary metabolites. For instance, higher antioxidant activity and phenolic content were observed in plants grown under specific climatic conditions. The composition of essential oils in *V. officinalis* can vary with seasons, indicating that climatic changes throughout the year affect the plant's chemical profile. While the influence of soil and climatic conditions is crucial, it is also important to consider genetic variability among different populations of *V.*

*officinalis*, which may contribute to differences in chemical composition independent of environmental factors (Letchamo *et al.* 2004; Heydari-Rahni *et al.* 2022; Raj *et al.* 2023).



**Fig. 1.** The factors that can affect *Valeriana officinalis* components include season, climate, pH of soil, harvested method, planting method, humidity, method of extracting and light.

The main objective of this research is to determine the most appropriate variety of *V. officinalis* by differentiating between thin- and thick-rooted variants. Furthermore, the study aims to analyze the effects of various plantation establishment techniques on the growth of underground structures, including roots and rhizomes. In addition, it intends to evaluate the yield results and the levels of essential oil and valerenic acid present in the roots and rhizomes, which are critical indicators of the quality of the raw materials.

### Literature Review and Selection

An extensive narrative review of the existing literature concerning the context of diverse elements can affect *V. officinalis* chemical contents. The criteria for inclusion comprised articles published in English, accessible in full-text format, comprehensive in scope, and directly pertinent to the research topic under examination. A meticulous search was performed in the PubMed and Scopus databases in December 2024, employing keywords related to *V. officinalis*, Valerian, soil, season, extraction, humidity, light, and component. The initial search yielded 149 articles based on their titles, abstracts, and publication dates. Following the removal of duplicate entries, 45 unique articles remained. The full texts of these articles were thoroughly reviewed, and a selection of three articles relevant to the research inquiry was identified. Subsequently, in December 2024, an additional search was conducted utilizing Google Scholar, PubMed, and Scopus, identifying and including three further articles that were directly relevant to the subject of interest. Nine additional references were incorporated throughout the writing process to improve the clarity and coherence of the arguments presented. To enhance the clarity and coherence of our arguments, nine additional references were integrated throughout the writing process.

### *Valeriana officinalis*

Valerian (*V. officinalis*, Valerianaceae) is a robust perennial flowering plant characterized by clusters of fragrant pink or white blossoms. In the northern hemisphere, these flowers typically bloom from June to September. Historically, during the 16<sup>th</sup> century, valerian was utilized as a perfume, and its natural distribution includes Europe and parts of Asia, with subsequent introduction to North America (Sharma *et al.* 2010; Ilyina *et al.* 2017). In pharmacology and phototherapy, valerian is an herb or dietary supplement derived from the plant's roots. This medicinal herb is widely recognized and has a long-standing reputation for its therapeutic benefits, particularly as a tranquilizer or sedative, making it especially beneficial for individuals experiencing nervous tension. Additionally, valerian is incorporated into perfumes to impart a mossy fragrance, although many often deem this

scent unpleasant. Research indicates that valerian promotes sleep, enhances sleep quality, antimicrobial effect, and lowers blood pressure. It is also employed in the management of conditions such as painful menstruation, cramps, hypertension, and irritable bowel syndrome. However, it is contraindicated for individuals with liver issues (Mohammadi *et al.* 2023, 2024). The active compounds, known as valepotriates, have been shown to exert a calming influence on agitated individuals while also acting as a stimulant in cases of fatigue (Ilyina *et al.* 2017; Dhiman *et al.* 2020; Heydari-Rahni *et al.* 2022). The phytochemistry of valerian include (Bos 1997; Nandhini *et al.* 2018):

- 1) Iridoid valepotriates (0.5% - 2.0%): including valtrate, isovaltrate, didrovaltrate, valerosidate, among others.
- 2) Volatile essential oil (0.2% - 2.8%): comprising bornyl isovalerate and bornyl acetate; valerenic, valeric, isovaleric, and acetoxvalerenic acids; valerenal, valeranone, cryptofaurinol; as well as various monoterpenes and sesquiterpenes.
- 3) Alkaloids (0.01% - 0.05%): valeranine, chatinine, alpha-methyl pyrrolketone, actinidine, skyanthine, and naphthylidyl methyl ketone.
- 4) Lignans: notably hydroxypinoresinol.

Valerian is known to contain over 150 distinct chemical constituents, many of which exhibit physiological activity. There is considerable variability in these constituents depending on factors such as the source of the plant, its growing conditions, processing techniques, and storage methods (Malhotra *et al.* 2024). Even standardized extracts available in Germany show some differences in the concentrations of various chemical components, which may influence their clinical effectiveness (Bauer 1998). Nevertheless, the therapeutic effects of valerian appear to be consistently observed across different formulations. While the sedative properties of valerian root have been recognized for centuries, the specific chemical compounds responsible for these effects remain unidentified and subject to debate. There is minimal correlation between the levels of volatile oils and the clinical outcomes associated with the plant. The central nervous system effects of valerian have been attributed to a range of constituents, including valepotriates, their metabolites (ballerinas), valerenic acid, valerenal, valeranone, and other components found in the essential oil (Haider *et al.* 2024; Malhotra *et al.* 2024). The presence of isovaleric acid contributes to the herb's distinctive and often unpleasant odor. Additionally, actinidine is a potent attractant for felines, known to engage with valerian, such as in their response to catnip. Furthermore, valerian is among several plant species that accumulate chromium, which may be utilized to address chromium deficiencies in certain developing regions (Sharma *et al.* 2010; Heydari-Rahni *et al.* 2022).

### **Effects of seasons and temperature on *V. officinalis* components**

Climatic conditions, including temperature, humidity, and light exposure, significantly impact the growth and development of *V. officinalis*. These factors can influence the plant's metabolic pathways, leading to variations in its chemical composition. Understanding the optimal climatic conditions for the *V. officinalis* cultivation is essential for producing high-quality medicinal products. Temperature fluctuations can significantly impact the chemical composition of plants, as they affect the plant's metabolic pathways. Temperature changes can influence the concentration of bioactive compounds, such as valerenic acid, and alter the plant's medicinal properties (Wu *et al.* 2023; Alum 2024). Understanding the effects of temperature fluctuations on plants is crucial for optimizing their cultivation. Light intensity can significantly impact the chemical composition of *V. officinalis*, as it affects the plant's photosynthetic pathways. Changes in light intensity can influence the production of bioactive compounds, such as valerenic acid, and alter the plant's medicinal properties. Understanding the effects of light intensity on *V. officinalis* is essential for optimizing its cultivation (Wills & Shohet 2009; Thakur *et al.* 2024).

In an experimental study, the roots of two-year-old plants are collected in the autumn after the foliage has withered, and they can be utilized either in their fresh state or after drying. Fresh roots exhibit approximately three times the efficacy compared to those dried at 40 °C. Furthermore, exposure to temperatures exceeding 82 °C results in the degradation of the active compounds present in the roots. The root yields approximately 1% essential oil. To maximize the production of commercial essential oil, many cultivators adopt high planting densities, typically ranging from 6 to 10 plants per square meter for annual crops. At the same time, this approach is not applied to biennial crops. Increased plant density may elevate relative humidity within the canopy and prolong leaf wetness by limiting air circulation and sunlight penetration (Morteza *et al.* 2009). Douglas *et al.* (1996) identified an optimal density of 6 plants m<sup>-2</sup> for maximizing essential oil yield in valerian. Additionally, most studies assessing the influence of plant density on valerian yield have historically employed analysis of variance

and mean separation methods to evaluate differences across various plant density levels. The correlation between essential oil yield and sowing date remains unestablished. Early sowing extends the duration that the plant is exposed to environmental conditions, which is also linked to a higher incidence of several diseases. Therefore, early planting may increase the likelihood of adverse effects, including the quality of the essential oil. Several studies have investigated the relationship between sowing date and essential oil yield (Morteza *et al.* 2009). Direct seeding, especially during the spring season, along with the conventional method of transplanting seedlings in spring, resulted in a notably reduced quantity of thinner roots and diminished raw material yields. Research conducted on this plant indicated that cultivating this species from seedlings grown in multi-cell trays led to the development of more branched roots with greater diameters, in contrast to plants propagated through direct seeding. This method consequently resulted in a marked increase in yields for those plants propagated from seedlings (Wiśniewski *et al.* 2016). Similar findings were reported by Kołodziej (2012) regarding artichoke cultivation, while Kołodziej & Najda (2007) reported that producing seedlings in multi-cell trays prolonged the growth period and positively influenced raw material yields, particularly for species vulnerable to low temperatures. Importantly, whether seeds were sown directly in the field or a seedbed, establishing plantations in the autumn resulted in thicker and heavier roots than those planted in spring. This phenomenon can be attributed to the fact that autumn seeding facilitated an earlier start to vegetative growth in spring, leading to higher yields than those achieved through spring seeding (Kołodziej & Najda 2007; Kołodziej 2012). The highest yields of roots and rhizomes were achieved with the thick-rooted variety, which was grown from seedlings that were initially produced in a seedbed and then transplanted in the autumn. Generally, autumn planting yielded more underground parts than spring planting. The discrepancies noted in this experiment may stem from the longer growth period of plants cultivated in multi-cell trays or those sown in seedbeds and transplanted in autumn compared to those planted in spring. Furthermore, the enhanced growth of seedlings in both seedbeds and multi-cell trays contributed to the increased raw material yields, as opposed to those obtained from direct field sowing. Farmers often face issues with lower germination rates of valerian seeds during spring sowing, as these seeds exhibit their highest viability immediately following harvest in August (Wiśniewski *et al.* 2016). Previous studies posited that variations in seasonal conditions and light exposure could influence the levels of medicinally beneficial components alongside alterations in the plant's morpho-physiological characteristics. Cultivating the plants under optimal light conditions and harvesting the appropriate plant parts during the ideal season is essential for maximizing the crop's potential. Throughout all seasons, growth metrics—including plant height, leaf count, leaf area, relative water content, and biomass—were found to be superior in shaded conditions. Conversely, total flavonoids, tannins, phenolic compounds, and antioxidant activities were elevated in full sunlight. High-performance liquid chromatography (HPLC) analysis indicated that valerenic acid and a majority of phenolic compounds peaked during the summer, particularly in the leaves. The findings advocate for harvesting *V. jatamansi*, especially the leaves, in the summer to obtain high-quality raw materials while minimizing the risk of depleting below-ground parts (Pandey *et al.* 2021). Regarding valerenic acid, the most maximal concentration of this compound was observed in valerian roots sourced from direct seed sowing and autumn seedling transplantation, as opposed to those derived from spring transplantation, which both seedlings were cultivated in multi-cell trays (Wiśniewski *et al.* 2016).

### **Effect of chemical fertilizers on *Valeriana officinalis* components**

Soil pH significantly influences the chemical composition of *V. officinalis*, as it affects the availability of essential nutrients and micronutrients. A specific pH range can alter the concentration of bioactive compounds, such as valerenic acid, in the plant. Understanding the optimal soil pH for *V. officinalis* cultivation is crucial for maximizing its medicinal properties. Soil nutrient availability, particularly nitrogen, phosphorus, and potassium, affects the chemical composition of plants. The plant's ability to absorb and utilize these nutrients influences the production of bioactive compounds, such as valerenic acid and isovaleric acid (Radanović *et al.* 2006; Adamczyk-Szabela *et al.* 2015). Optimizing soil nutrient levels can enhance the medicinal properties of *V. officinalis*. Cultivation practices, including irrigation, fertilization, and pruning, can significantly impact the chemical composition of plants. Optimizing cultivation practices can enhance the production of bioactive compounds, such as valerenic acid, and improve the plant's medicinal properties (Heydari-Rahni *et al.* 2022). This research investigated the impact of urea-based chemical fertilizers applied at five different levels alongside four types of biofertilizers (Nitroxin, Phosphate Barvar2, a combination of both, and a control group) on the yield quantity and

quality of valerian from 2016 to 2020. The parameters assessed include the percentage and total grams per hectare of valerenic acid, root dry weight, shoot dry weight, root diameter (mm), root length (cm), leaf width (cm), and leaf length (cm). The findings indicated that biofertilizers significantly influenced all measured quantity and quality parameters. In contrast, all levels of urea fertilizer negatively affected both the yield and the percentage of valerenic acid, while biofertilizers enhanced these metrics. The highest percentage of valerenic acid was recorded in the control group, while the lowest was observed at the 150 kg ha<sup>-1</sup> urea level. Compared to the control group, the greatest root dry weight was associated with the combination of Nitroxin, Phosphate Barvar2, and 150 kg urea (Heydari-Rahni *et al.* 2022). These results suggest that biofertilizers, which exhibited beneficial effects on the studied parameters, could serve as viable alternatives to chemical fertilizers. Mikovacki & Milic reported that the application of Nitroxin biofertilizer notably influenced root length. Their findings indicated that both the individual and combined use of nitrogen and phosphorus biofertilizers enhanced the roots' capacity to absorb nutrients, leading to a favorable increase in yield parameters due to the enhancement of the root system (Mrkovacki & Milic 2001). In agreement with reported studies, it was indicated that the simultaneous use of nitrogen and phosphorus biofertilizers alongside inorganic fertilizers yields superior results in terms of both yield and yield components of the target plants compared to their applications. Consequently, the integrated application of biofertilizers with inorganic fertilizers demonstrates greater efficacy than their standalone use, attributed to beneficial interactions among their microorganisms, ultimately leading to enhanced yield components (Mrkovacki & Milic 2001). A study was conducted to investigate the influence of Myconate® HB (formononetin) on the growth parameters, biomass, nutrient composition, and bioactive compounds in *V. officinalis* and *Salvia officinalis*. Findings indicated that the application of Myconate® on *V. officinalis* seedlings resulted in a beneficial increase in both the biomass of aerial parts and root systems, alongside enhancements in dry matter and fiber content. Conversely, the levels of total protein and nitrogen-free extracts were found to be less favorable. Notably, Myconate® did not influence the duration of the phenological stages in the plants under study. Throughout the growing season, the percentage of essential oils increased in both species across all experimental plots, with significantly higher concentrations observed from the full tillering stage in the plots treated with Myconate® (Mrkovacki & Milic 2001). The experiment conducted by Domenico Prisa & Giordano Menci demonstrated that applying innovative fertilizers infused with plant extracts can substantially enhance the vegetative and root development of *V. officinalis* L. and *Raphanus sativus*. All experimental treatments with fertilizers exhibited a marked improvement compared to the untreated control and the commercial treatment with *Ecklonia maxima* concerning the agronomic parameters assessed. Notably, the Qi-gen treatment emerged as the most effective in promoting vegetative and root biomass increases. Additionally, plant height, leaf count, and root length enhancements were observed. The trial further indicated a significant impact of the product Aktigen on controlling *Botrytis* (a fungus), as well as the product Lifegen on managing powdery mildew, both of which effectively reduced the mortality rates of seedlings of *V. officinalis* and *R. sativus* (Mrkovacki & Milic 2001).

### **Effect of extraction methods on valerian components**

The methods employed for extraction play a crucial role in determining valerian constituents' composition and effectiveness. Various solvents and techniques result in differing concentrations of bioactive compounds, which in turn impacts their possible therapeutic applications. Ethanol and methanol are frequently utilized as solvents in the extraction of valerian, each presenting unique benefits regarding compound yield and bioactivity. Consequently, the selection of an extraction method can be strategically aligned to enhance the properties of valerian extracts for particular applications (Mrkovacki & Milic 2001). To enhance the extraction yield of bioactive compounds from the roots of *V. officinalis*, a D-optimal design based on response surface methodology was employed by Mokhtari *et al.* (2024). This approach involved investigating the influence of several variables, including the type and concentration of solvent, mixing temperature, duration of ultrasound treatment, and drying technique. The optimal parameters identified were a solvent concentration of 94.88%, a mixing temperature of 25 °C, an ultrasound duration of 48.95 minutes, the use of methanol as the solvent, and microwave drying, achieving a desirability score of 0.921. Under the established optimal conditions, the actual yields of valerenic acid, total phenols, total flavonoids, and antioxidant activity were measured at 2.07 mg g<sup>-1</sup> DW, 7.96 mg g<sup>-1</sup> DW, 5.52 mg g<sup>-1</sup> DW, and 78.68%. This research serves as a valuable framework for illustrating the effectiveness of microwave drying in maximizing the biochemical constituents of *V. officinalis* and enhancing the antioxidant properties of its root extracts for industrial applications (Mokhtari *et al.* 2024). Valerian, *V. officinalis* is a medicinal plant

indigenous to Europe and North Asia, with its roots and rhizomes utilized for their sedative properties in alleviating nervous tension, insomnia, anxiety, and stress. A study was conducted to assess the effects of washing and drying on the concentration of valerenic acids in valerian roots. The analysis revealed that the valerenic acid content in roots subjected to washing, soaking, or cutting before drying did not significantly differ from that of whole valerian roots dried without prior washing. However, cutting the roots notably reduced the drying duration. Increasing the drying temperature from 15 to 70 °C reduced drying time by 176 hours, accompanied by a 23% loss of valerenic acids between 15 and 40 °C. An additional 36% loss was observed when the temperature was raised from 40 to 50 °C, while no significant changes were noted at higher temperatures. The application of a heat pump dryer at 40 °C decreased the drying time by 25% compared to a conventional hot air dryer at the same temperature, with no significant difference in valerenic acid levels. Furthermore, fresh whole roots stored at ambient conditions for 10 days exhibited no significant change in valerenic acid levels, although a notable reduction in water content was recorded. Consequently, valerian cultivators may opt to separate or soak roots before washing and can store fresh roots before drying without significant concern regarding valerenic acid loss. Nonetheless, it is advisable to dry the roots at the lowest feasible temperature (Shohet & Wills 2006; Shohet & Wills 2006; Wills & Shohet 2009). Valerian extracts derived from its underground parts, specifically the roots and rhizomes, have been used for medicinal purposes for over a century. Certain valerian extracts, particularly aqueous ones, have been recognized for their sedative and anxiolytic properties. Nevertheless, the precise identification of the active constituents needs to be clarified. Traditional methods such as hydro distillation and steam distillation have been employed for the recovery of essential oils from plant materials; however, these methods present several drawbacks, including low yield, prolonged distillation times, loss of volatile compounds, and high operating temperatures that may lead to the degradation of the essential oil's chemical constituents. Furthermore, there are limited adjustable parameters in hydro distillation, steam distillation, and solvent extraction that can influence the selectivity of the extraction process. As a result, there is a pressing need for the development of more efficient extraction techniques with enhanced selectivity. In response to this need, supercritical fluid extraction (SFE) has been introduced, and extensive research has been done on separating active compounds from various herbs and plants. SFE is an effective method for producing flavors and fragrances from natural sources and serves as a viable alternative to the traditional essential oil extraction techniques. This method offers a rapid and quantitative approach to extracting essential oils from aromatic plants. By optimizing the conditions of SFE, such as pressure and temperature, it is possible to manipulate the density of the supercritical fluid, thereby influencing its solvent capabilities. Research has been conducted on the supercritical CO<sub>2</sub> extraction of the valerian, *V. officinalis* L. roots (Zizovic *et al.* 2007). Based on previous studies, when extracted using 95% ethanol at a temperature of 75 °C, valerian roots and rhizomes exhibited the highest concentrations of bioactive compounds, notably valerenic acid derivatives and phenolic acids. The resulting extracts displayed enhanced antioxidant properties and effectively inhibited enzymes associated with metabolic syndrome, including pancreatic lipase and angiotensin-converting enzyme (ACE). On the other side, methanol tinctures of valerian roots identified 77 components, with some compounds present in higher quantities compared to ethanol extracts. Methanol extraction was noted for yielding a diverse range of components, although ethanol provided better quantitative and qualitative composition (Wu *et al.* 2023). Ethanol and methanol extractions are known to produce highly effective bioactive compounds; however, the selection of solvent and extraction method significantly influences the stability and selectivity of the resulting extracts. For instance, hydro alcoholic extracts exhibited distinct interactions with glutamate receptors in contrast to aqueous extracts, underscoring the critical role of solvent selection in shaping the characteristics of the extracts. Additionally, the potential for drug interactions and the necessity to thoroughly evaluate valerian's pharmacokinetic properties are vital considerations when incorporating these extracts into treatment protocols. Furthermore, pulsed ultrasound-assisted extraction has demonstrated superior efficiency compared to traditional methods, resulting in an increased yield of bioactive compounds from valerian roots. This technique effectively disrupts cell walls, thereby enhancing the extractability of both readily available and more challenging polysaccharides (Sultana *et al.* 2009). The study by Negi *et al.* examined the impact of various extracting solvents, specifically dichloromethane, ethyl acetate, and methanol, on the valerenic acid concentration and antioxidant properties of root extracts from both cultivated and wild *Valeriana jatamansi*. Notably, dichloromethane yielded higher concentrations of valerenic acid and exhibited superior antioxidant activity compared to ethyl acetate and methanol. The findings indicate that the level of valerenic acid is crucial for the antioxidant efficacy of *V. jatamansi*. The hierarchy of valerenic acid concentration

across different samples was established as follows: dichloromethane > ethyl acetate > methanol. This pattern was similarly observed in the antioxidant activity of the plant extracts (Negi *et al.* 2012). Currently, microwave-assisted extraction technology is extensively employed for the extraction of essential oils. This technique enables the extraction process to be completed briefly, thereby minimizing pyrolysis, hydrolysis, and oxidation occurrences. The findings of the experimental study indicated that the ideal conditions for the microwave extraction of volatile oil from valerian were established at a microwave power of 350W, an extraction duration of 25 minutes, and a liquid-to-material ratio of 8.8 mL to 1.0 g. Under these parameters, the yield of volatile oil obtained was 5.88%. The volatile oil extracted via microwave demonstrated significant scavenging activity and potent reducing capacity against DPPH and hydroxyl free radicals. When compared to steam distillation, notable differences were observed, which may serve as a valuable reference for the advancement of antioxidant products derived from valerian oil (Miao *et al.* 2020). Based on a previous study carried out by Hosseini Nia *et al.* in (2019), the dried root was subjected to extraction using three distinct methods: maceration, ultrasonic-assisted extraction, and Soxhlet extraction. The ultrasonic-assisted extract exhibited the highest levels of total phenolics and flavonoids. In terms of DPPH radical scavenging activity and reducing power, the ultrasonic-assisted extract demonstrated superior activity compared to the other extraction methods. Specifically, the IC<sub>50</sub> values for the ultrasonic, Soxhlet, and maceration extracts were 0.546, 0.816, and 0.678 mg mL<sup>-1</sup>, respectively. These findings indicate that the extraction methods employed in this study significantly influenced the antioxidant capacities and the total phenolic and flavonoid contents. It was determined that both ultrasonic-assisted and Soxhlet extraction methods are more effective for extracting antioxidant components from *V. officinalis* L.

### **Effect of density planting**

The findings indicated that delayed planting adversely impacted valerian's quantity and quality attributes. The timing of valerian planting is a critical management choice for producing essential oils. The selection of planting dates influences the incidence of leaf spots by mitigating unfavorable weather conditions that promote disease. A later planting date correlated positively with increased necrosis due to more favorable weather conditions. Conversely, early planting enhances the potential for forage production by prolonging the vegetative growth phase, thereby increasing the duration that valerian is exposed to environmental factors. In regions with limited soil moisture, premature planting may lead to excessive growth in the fall, resulting in the depletion of soil moisture necessary for early spring growth (Morteza *et al.* 2009; Raj *et al.* 2024). Additionally, early planting of valerian can lead to an earlier break from winter dormancy as temperatures rise, which may increase the risk of late spring freeze damage. Valerian planted at an intermediate date demonstrates a higher potential for essential oil yield compared to late-planted valerian, attributed to the development of more lateral stems, leaves, and flowering stems. Late-planted valerian grows under different temperatures and photoperiod conditions, experiences a reduced vegetative growth period, and necessitates a higher essential oil yield to offset the diminished development of flowering partially stems (Morteza *et al.* 2009). The study analyzed five wild populations of valerian from *Latvia* in which the concentration of essential oil in samples collected during the autumn 2017 ranged from 8.4 to 10.7 mL kg<sup>-1</sup>, while the subsequent spring harvest in 2018 exhibited a higher essential oil concentration of 11.5 to 14.2 mL kg<sup>-1</sup>. According to the European Pharmacopoeia (EP) standards, the total essential oil content in the dried valerian roots should exceed 4 mL kg<sup>-1</sup>, and all examined wild populations satisfied this requirement. This research represents the first documentation of essential oil content in wild-collected valerian roots from *Latvia* (Nakurte *et al.* 2021). Prior investigations conducted in Estonia and analyses of commercial samples from pharmacies and health stores across various countries—including Belgium, the Czech Republic, France, Germany, Greece, Hungary, Latvia, Lithuania, Moldova, Russia, the United Kingdom (Scotland), and Ukraine—indicated that the essential oil content in valerian roots ranged from 0.19% to 1.16%. In Lithuania, cultivated valerian samples exhibited an essential oil content of 0.55%, while roots collected in Bulgaria yielded 0.40-0.42% (Baranauskiene 2007). A study of wild populations in Hungary reported essential oil content varying from 0.32 mL 100 kg<sup>-1</sup> (3.2 mL kg<sup>-1</sup>) to 0.60 mL 100 kg<sup>-1</sup> (6.0 mL kg<sup>-1</sup>; Sárosi *et al.* 2005). Additionally, wild roots harvested from mountainous regions in Serbia produced an essential oil content of 1.88% (Pavlovic *et al.* 2004). The observed variation in essential oil content across different growth phases and harvesting times aligns with findings from previous studies; for instance, valerian cultivated in Poland showed oil content ranging from 0.8% at the beginning of vegetation to 1.3% at full bloom and from 0.75% after the first frost to 1.25% three weeks post-seed harvest (Seidler-Lozykowska *et al.* 2009). A study (Morteza *et al.* 2009)



examined three different planting dates (March 5, March 15, and March 25) alongside three varying plant densities (50 × 20 cm, 50 × 30 cm, and 50 × 40 cm). Findings indicated that the earliest planting date (March 5) yielded the highest metrics in terms of flower count per plant, fresh and dried flower weight per plant, as well as fresh flower yield, dried flower yield, essential oil yield, chamazulene percentage, and chamazulene yield. Furthermore, the lowest plant density (50 × 40 cm) resulted in the greatest number of fresh flowers per plant and dried flowers per plant. Conversely, the highest fresh, dried, essential oil, and chamazulene yields were recorded at the highest plant density (50 × 20 cm). Thus, the investigation concluded that optimal yield is achieved with early sowing and higher plant density. Additionally, the maximum root yield was observed at an optimal plant density of 8 plants m<sup>2</sup>, attributed to enhanced photosynthesis resulting from increased leaf area, which in turn elevates essential oil content. Plant population density significantly influences both vegetative and reproductive growth in valerian. Low plant density correlates with reduced valerian yield due to limited leaf area plasticity per plant. Moreover, valerian exhibits a limited ability to form new reproductive structures in response to increased resource availability per plant (Morteza *et al.* 2009). The treatment involving a planting density of 4 plants per meter in a single row yielded the highest quality of marketable and exportable produce. Conversely, the lowest marketable yield was recorded at the highest plant density. The critical threshold for plant density was identified at 0.2 m with a single-row configuration. Increased plant density has been shown to enhance the essential oil content in species such as sweet annie and cumin. However, excessively high plant density can reduce resource availability per individual plant during the flowering stage, resulting in a significant decline in yield per plant that is not compensated by the increase in plant numbers. Additionally, high plant density can elevate relative humidity within the canopy and prolong leaf wetness due to diminished air circulation and sunlight penetration. In response to low-density conditions, plants may increase their shoot growth to enhance photosynthetic efficiency, which in turn leads to an elevation in root length at a planting density of 12 plants per square meter (Carrasco *et al.* 1996). The number of flowering stems was increased in low-density environments, where diminished competition among plants led to a rise in the reproductive cells associated with flowering. In high-density situations, plants tended to elongate their roots to improve water uptake, driven by increased competition. However, in low-density settings, the diameter of the roots increases, which can be linked to adequate assimilation processes that promote root expansion. Consequently, it is evident that plant density significantly affects the occurrence of plant diseases (Douglas *et al.* 1995). An investigation (Tabatabaei 2008) into the growth and essential oil production of *V. officinalis* L. utilized various cultivation systems, including aeroponic, floating, growing media, and soil. The floating media system yielded the highest fresh weights for both leaves and roots. While no significant differences in leaf area were noted between the floating and growing media systems, a marked reduction in leaf area was observed in the aeroponic and soil systems. Additionally, both photosynthesis rates and stomatal conductance were enhanced in the floating and growing media systems compared to the aeroponic and soil systems, which also correlated with the increased essential oil concentration. The primary components of the essential oil included bornyl acetate, vale-renal, camphene, trans-caryophyllene, cis-ocimene,  $\alpha$ -fenchone, and  $\delta$ -elemente, with variations in their relative proportions depending on the treatment. Notably, the concentration of bornyl acetate reached its peak at 32.1% of the total oil in the floating system, representing a 56.5% increase over the soil system. Furthermore, the soil system promoted the production of valerenal, which was found in higher concentrations compared to the other growth systems. These findings indicate that both floating and growing media systems may effectively enhance root yields and essential oil production in valerian under controlled environmental conditions (Tabatabaei 2008).

## CONCLUSION

Based on the present study, the pH level and nutrient availability in the soil profoundly impact the uptake of vital nutrients, subsequently altering the levels of bioactive compounds such as valerenic acid. This underscores the critical role of soil management practices in the cultivation of high-quality valerian plants. Climatic elements, including temperature, humidity, and light exposure, are essential in the biochemical processes of *Valeriana officinalis*. These environmental factors can result in notable variations in the plant's chemical composition, thereby influencing its medicinal attributes. Essential macronutrients, such as nitrogen, phosphorus, and potassium, are crucial for the production of beneficial compounds in valerian. Maintaining sufficient levels of these nutrients is necessary to enhance the plant's therapeutic potential. Variations in temperature and water supply can trigger the production of compounds associated with stress, which may modify the plant's therapeutic characteristics. This indicates that environmental stressors can exert both beneficial and detrimental effects on the

chemical profile of valerian. The study concludes that optimizing both soil conditions and climatic factors could significantly enhance the medicinal properties of *V. officinalis*. This highlights the importance of implementing meticulous cultivation strategies to improve the quality of valerian as a medicinal herb. The research emphasizes the significant influence of soil and climatic conditions on the chemical composition of *V. officinalis*, which affects its growth and phytochemical profiles. By examining these factors, the study aims to provide insights into how variations in environmental conditions can lead to differences in the concentration of essential oils, phenolic compounds, and other bioactive constituents. Ultimately, the goal is to enhance the understanding of optimal cultivation practices that can improve the quality and yield of this medicinal plant, ensuring that it meets the stringent requirements of the pharmaceutical industry.

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