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# Impact of Agrotechnical Methods on the Development and Economic Indicators of *Elsholtzia ciliata* (Thunb.) Hyl.

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Elsholtzia ciliata (Thunb.) Hyl., commonly known as Vietnamese balm, is an aromatic and medicinal plant valued for its essential oil rich in biologically active compounds. This study aimed to evaluate the influence of sowing time and irrigation regime on the growth, productivity, and essential oil content of E. ciliata genotype No. 23–60 cultivated under the climatic conditions of Cherkasy Oblast, Ukraine. Field experiments included phenological observations, morphometric measurements, and assessment of biomass and essential oil yield under three watering regimes (natural, moderate, and intensive drip irrigation) for two sowing dates. The total vegetative period was 144 days for the first sowing and 131 days for the second. Plants grown under intensive watering showed enhanced growth parameters and higher biomass production compared with those under natural irrigation. The highest green mass and inflorescence yields were recorded at the first sowing with intensive drip irrigation, while moderate irrigation provided the maximal leaf yield. Essential oil content in the mass-flowering phase ranged from 0.45–0.82% for the first sowing and from 0.57–0.59% for the second, depending on the water regime. The results indicate that early sowing combined with intensive drip irrigation ensures optimal growth and oil accumulation in E. ciliata. These findings may be useful for developing agrotechnical recommendations and for further breeding aimed at selecting genotypes with improved essential oil yield and adaptability to different environmental conditions.

Keywords: Vietnamese balm, sowing date, irrigation regime, phenology, biomass yield, essential oil content

# Introduction

The importance of aromatic and medicinal plants as sources of biologically active substances is increasing. The increasing demand for bioactive compounds can be met by expanding the cultivation of medicinal and aromatic plants. Successful introduction of promising plant species into culture is possible only if their developmental biology is known, the processes of

formation and accumulation of essential oil are studied, and the features of their cultivation and productivity are studied to determine the feasibility of industrial cultivation (Belouri et al., 2022). One of the most widespread plant families that includes aromatic plants is Lamiaceae.

The genus *Elsholtzia* Willd. includes 48 species of the family Lamiaceae, from which 43 are found and

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used in China (Sahu and Sarankar, 2025). In the wild, the plants are found in East Asia, Africa, North America, and European countries, and are especially common in China, Japan, Korea, and India (Liu et al., 2007; Zhang et al., 2021). Most species of the genus grow at an altitude of 1000–3000 m. These are mainly aromatic plants that have been used in home folk medicine, as food, in cosmetics, and as a source of honey production. Another important property of plants is the restoration of soil from heavy metal pollution (Guo et al., 2012; Wang et al., 2022).

Representatives of the genus *Elsholtzia*, such as *E. patrinii* (Lepech.) Garcke, *E. ciliata* (Thunb.) Hyl. and *E. stauntonii* Benth. are distributed mainly in the temperate regions of the Northern Hemisphere, as well as in North America. They grow on hilly meadows, wastelands, in forests, and in valleys. They are cultivated in many countries of Europe and North America (Zotsenko, 2021; Svydenko, 2024). In Ukraine, *E. ciliata* is an introduced species, which is also found in the wild, in particular in Transcarpathia and Polesie. At the same time, it is not used in official medicine (Zotsenko and Tsukkan, 2017).

The plant raw material of *E. ciliata* exhibits antimicrobial, antispasmodic, expectorant, diaphoretic, hemostatic, analgesic, carminative, and diuretic effects (Guo et al., 2012; Zotsenko, 2021). In the traditional medicine of different countries of the world, it is used for the treatment of tuberculosis, edema, acute gastritis, and ascites, for the treatment of respiratory diseases, and jaundice. In addition, Korean scientists have investigated the possibility of using *Elsholtzia ciliata* essential oil for the treatment of drug addiction (Zhaoa et al., 2016). *Elsholtzia ciliata* essential oil *in vitro* showed antiproliferative activity against cancer cells (Pudziuvelyte et al., 2017).

This plant is a valuable source of biologically active substances (Liu et al., 2007; Shangzhen et al., 2009; Guo et al., 2012; Pudziuvelyte et al., 2018), among which are flavonoids, phenylpropanoids, terpenoids, and other compounds (Guo et al., 2012; Pudziuvelyte et al., 2020). *E. ciliata* is a valuable bioactive source of natural antioxidants (Liu et al., 2012; Ma et al., 2018; Nguen et al., 2021). The main components identified in the aerial parts were carvone, limonene, and  $\alpha$ -caryophyllene (Liang et al., 2020).

The essential oil is contained in all organs of the plant and contains 58 different components (Tian, 2013; Wang et al., 2017) and exhibits antimicrobial, insecticidal effects (Liang et al., 2020). Contact toxicity and fumigant activity against food pests have been

established for the essential oil from the aerial parts of *Elsholtzia ciliata* (Liang et al., 2020; Shen et al., 2024). It is an ornamental plant due to its bright, purple flowers and pleasant aroma, which is simultaneously reminiscent of the smell of citrus fruits, mint, and lemon balm. *Elsholtzia* is a good honey plant. Its honey productivity is up to 170 kg per hectare (Guo et al., 2012; Zotsenko and Tsurkan, 2017).

The rather wide use of the herb and essential oil of *Elsholtzia ciliata* dictates the study of possible areas for its cultivation and agricultural techniques. The plants had significantly greater height, leaf length, leaf width, and weight of the above-ground mass, and the highest essential oil content using gibberellic acid was 1.41% (Luong et al., 2024).

Most published studies on this species focus on its chemical composition and medical applications, with less information available on agrotechnical methods for growing *Elsholtzia ciliata*. However, many scientists have worked on improving the cultivation technology of other aromatic plants of the Lamiaceae family: for example, *Agastache foeniculum* (Pursh) Kuntze is highly sensitive to watering and soil fertilisation (Omidbaigi et al., 2008). It is noted that the yield and composition of essential oil in *A. foeniculum* are influenced by sowing dates and irrigation regime, with the best results obtained from early spring sowing (Omidbaigi and Sefidkon, 2004; Vârban et al., 2022).

Studies on the irrigation effect on peppermint yield in California have shown that oil yield in plants decreases with a delay in the start of irrigation (Marcum et al., 2006). In the arid conditions of Khorasan (Iran), the high potential of water saving was determined for *Thymus vulgaris* L. and *Hyssopus officinalis* L. through longer irrigation intervals (e.g., 14 days) (Khazaie et al., 2008).

The watering intervals affected on *Origanum vulgare* L. morphological characteristics except for the proportion of stems. With an increase in manure level, the number of stems, plant diameter, stem diameter, leaf area, and fresh and dry grass yield increased (Gerami et al., 2016). It has been proven that sowing dates have a significant impact on the growth and development of *Dracocephalum moldavica* L., the maximum yield and essential oil content of which were obtained during the first sowing period using the seedling method of cultivation (Okhchlar et al., 2012).

Although *E. ciliata* has been widely studied for its chemical composition and pharmacological properties, little is known about its agronomic performance under different environmental and irrigation conditions.

In Ukraine, data on its cultivation and productivity are scarce. Therefore, this study aimed to assess the influence of sowing time and irrigation regime on growth, biomass yield, and essential oil content of *E. ciliata* under the conditions of the Central Forest-Steppe of Ukraine.

# **Material and Methodology**

# Conditions of growth and plant material

The research was conducted in 2024 in the conditions of the Central Forest-Steppe of Ukraine in the Lysyansky district of the Cherkasy region. The climate of the Central Forest-Steppe is temperate continental, with relatively mild, little-snowy winters and warm, moderately humid summers. In the region of study, the total precipitation was 592.3 mm in 2024. The average air temperature for the year was +11.5 °C. In the summer months, it ranged from +10.3 to +37 °C, with an average of 22.8 °C. The area is located on the Dnieper Upland of the East European Plain in the basin of the Gnyly Tikich River. The soil of the area where the research was conducted is typical black soil. The soil is heavy loam in terms of granulometric composition. The humus content in the arable layer is 4.58% (Sytnyk and Trohymenko, 2013).

The plant raw material of *Elsholtzia ciliata* (Thunb.) Hyl. genotype 23–60 was studied. Seeds collected from local reproduction. Plant seeds were sown in two rows with a row spacing of 50 cm. The experimental plots were 10 m<sup>2</sup> and randomly assigned. The 4-fold repetition of the experiment was used.

The first sowing period was carried out on the 26<sup>th</sup> of April, and the second sowing period was carried out on the 6<sup>th</sup> of May. Plant irrigation was carried out in three variants, which included intensive drip irrigation, moderate drip irrigation, and natural moistening.

# Phenological Observations and Biometric Measurements

Phenological development was recorded according to the BBCH scale (Meier, 2018), covering the principal growth stages from sprouting (PGS-0) to senescence (PGS-9): PGS-0 (sprouting), PGS-2 (tillering), PGS-5 (Inflorescence emergence (main shoot), 51– inflorescence of flower buds is visible, 55 – half of inflorescence emerged), PGS-6 (flowering, 61 – beginning of flowering, 69 – end of flowering), PGS – 8

(ripening of maturity of fruit and seed, 81 – beginning of ripening of fruit colouration, 89 – fully ripe: fruit shows fully-ripecolour, beginning of fruit abscission), and PGS-9 (senescence, beginning of dormancy, 93 – beginning of leaf fall). The measurements were carried out on 10 plants. The appearance of the investigated plants during the vegetative period is represented in Figure 1.

The data on the start and end of the investigated growth stages were fixed depending on the irrigation type. Some flowering peculiarities were described using recommendations of Beabien and Johnson (1994). Biometric measurements were carried out every two weeks during the growing season of the plants, and the height and diameter of the plants.

#### **Determination of Essential Oil Content**

Samples of above-ground biomass were collected every two weeks during the growing season. In each experimental variant, plants were cut at a uniform height above the ground level. The mass fraction of essential oil in the samples was determined according to the Ginsberg method using a Clevenger-type apparatus (Elyemni et al., 2019).

For each determination, 100 g of freshly cut, aboveground plant material was finely crushed and placed into a round-bottom flask. The sample was then mixed with 300-400 mL of distilled water. The flask was sealed with a cork stopper fitted with the lower end of a condenser and heated on an electric stove. The mixture of essential oil and water vapors rose through the condenser, where it was cooled and condensed. The condensate flowed into a graduated receiver, in which the essential oil separated as a distinct layer above the water. Excess water was drained through the side tube. Distillation continued for approximately one hour, until no further separation of oil was observed. The essential oil vield was calculated as the percentage of essential oil volume relative to the fresh mass of the plant material.

# Statistical analysis

All data were expressed as mean ± standard deviation (SD) of three replicates. Statistical significance was evaluated by one-way ANOVA, and means were compared using Tukey's post-hoc test at p <0.05. Data processing was carried out using Statistica 13.3 software (TIBCO Inc., USA).



Figure 1 The main growth stages of *Elsholtzia ciliata* (Thunb.) Hyl. during the vegetation period A – tillering (PGS-2), B – stem elongation (PGS-3), C – development of vegetatively propagated organs (main shoot) (PGS-4), D, E – inflorescence emergence (main shoots) (PGS-5), F – flowering (PGS-5), G – development of fruit (PGS-7)

# **Results and Discussion**

Elsholtzia ciliata is one of the three species of the Elsholtzia genus found in Ukraine, together with the cultivated *E. stauntonii* Benth. and *E. densa* Benth. (Zotsenko et al., 2021). The conducting of phenological observations was one of the main tasks of this study. They are an essential part and key component of this study, due to crucial insights into plant growth dynamics, functionality of plant communities, and diversity (Fenner, 1998; Meier et al., 2009). Such observations are especially important for assessing the adaptation of plants introduced to new environmental conditions, when flowering duration and synchrony directly affect successful pollination and reproduction (Arroyo et al., 2021).

Spring 2024 in the Cherkasy region was characterized by high temperature variability – typical for the Forest-Steppe zone – with alternating cool and warm periods, and excessive early moisture followed by a drier late spring. Seedlings of *E. ciliata* appeared on the 7–8<sup>th</sup> day after the first sowing (26 April) and on the 6<sup>th</sup> day after the second sowing (6 May). Irrigation regime did not significantly influence sprouting time (Table 1).

After 20 days from emergence, the tillering phase (PGS-2) was observed in plants sown in the first term, whereas in the second term sowing, this phase commenced slightly earlier, after 17 days. By early July, 44 days following the onset of tillering, plants from the first sowing term entered the budding stage (PGS-5), while in the second sowing, budding began after a slightly shorter interval of 42 days. These results indicate that later sowing can accelerate early vegetative development, possibly due to higher

temperatures and soil moisture levels during late spring, which favor faster shoot elongation.

Flowering phenology is a critical factor in understanding plant reproductive strategies and is strongly influenced by seasonal climatic conditions (Martins Eburneo et al., 2021). In this study, the interval from budding to the onset of flowering was 3-4 days shorter in plants sown in the second term, suggesting a compressed reproductive phase in later-sown plants. The beginning of flowering occurred at the start of August in both sowing periods. Notably, plants from the first sowing bloomed uniformly across all irrigation treatments, whereas in the second sowing, those under intensive and moderate watering initiated flowering slightly earlier. The termination of flowering in the second sowing occurred 2-3 days earlier than in the first, which may reflect environmental influences on reproductive timing, such as temperature fluctuations and water availability.

The total vegetative period is generally regulated by growth and climatic factors, including temperature and water regimes (Liubchenko et al., 2020), while the development of specific plant organs is controlled at the molecular level (Poething and Fouracre, 2024). In Cherkasy Oblast, the overall vegetative period lasted 142 days for the first sowing and 131 days for the second sowing (withoutirrigation). The summarized vegetative period with separated growth phases of *E. ciliata* is represented in Figure 2. The vegetative period can appear longer than actual organ development due to the overlap of growth stages; for instance, flowering may begin before stem elongation is complete. Detailed analysis of phase durations revealed that for the first sowing, the interval from sprouting to tillering lasted

Table 1 The phenological data of *Elsholtzia ciliata* (Thunb.) Hyl genotype 23–60 depends on sowing dates and irrigation

| Phenological<br>stages | 1 <sup>st</sup> term (26. 04.) |                             |                            | 2 <sup>nd</sup> term (06. 05.) |                             |                            |  |
|------------------------|--------------------------------|-----------------------------|----------------------------|--------------------------------|-----------------------------|----------------------------|--|
|                        | intensive drip<br>irrigation   | moderate drip<br>irrigation | natural drip<br>irrigation | intensive drip<br>irrigation   | moderate drip<br>irrigation | natural drip<br>irrigation |  |
| PGS-0                  | 02. 05. 24                     | 02. 05. 24                  | 02. 05. 24                 | 12.05.24                       | 12. 05. 24                  | 12. 05. 24                 |  |
| PGS-2                  | 22.05.24                       | 22. 05. 24                  | 22. 05. 24                 | 29. 06. 24                     | 29. 06. 24                  | 29. 05. 24                 |  |
| PGS-5, 51              | 06.07.24                       | 06. 07. 24                  | 06. 07. 24                 | 12.07.24                       | 12.07.24                    | 12.07.24                   |  |
| PGS-5, 55              | 10.08.24                       | 10.08.24                    | 08. 08. 24                 | 10.08.24                       | 10.08.24                    | 08. 08. 24                 |  |
| PGS-6, 61              | 09. 08. 24                     | 09. 08. 24                  | 06. 08. 24                 | 08. 08. 24                     | 08. 09. 24                  | 07. 08. 24                 |  |
| PGS-6, 65              | 17. 08. 24                     | 17. 08. 24                  | 15. 08. 24                 | 19. 08. 24                     | 19. 08. 24                  | 17. 08. 24                 |  |
| PGS-6, 69              | 05. 09. 24                     | 05. 09. 24                  | 04. 09. 24                 | 07. 09. 24                     | 07. 09. 24                  | 05. 09. 24                 |  |
| PGS-8, 81              | 23. 08. 24                     | 23. 08. 24                  | 22. 08. 24                 | 25. 08. 24                     | 25. 08. 24                  | 23. 08. 24                 |  |
| PGS-8, 89              | 03.09.24                       | 03. 09. 24                  | 01. 09. 24                 | 04.09.24                       | 04. 09. 24                  | 02. 09. 24                 |  |
| PGS-9                  | 22. 09. 24                     | 22. 09. 24                  | 20. 09. 24                 | 24. 09. 24                     | 24. 09. 24                  | 19. 09. 24                 |  |

Note: PGS – principal growth stage (according to BBCH-sale)

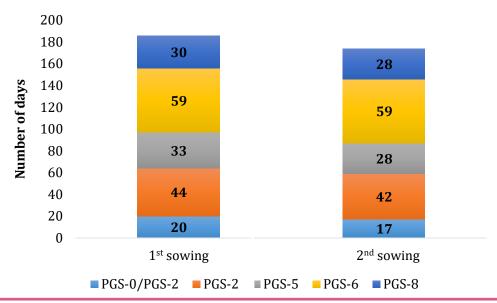
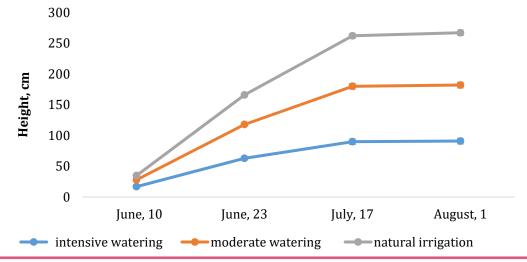


Figure 2 The vegetative period duration of *Elsholtzia ciliata* (Thunb.) Hyl. genotype 23–60 depends on sowing PGS – principal growth stage (according to BBCH-scale)

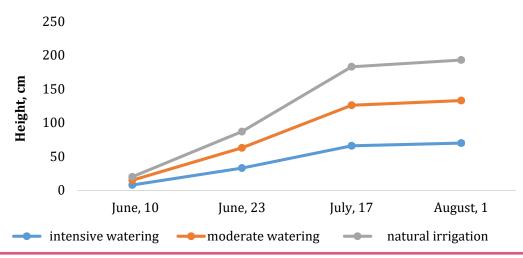
20 days, tillering 44 days, inflorescence emergence 33 days, flowering 59 days, and ripening 30 days. In the second sowing, the corresponding durations were 17, 42, 28, 59, and 28 days, respectively. Interestingly, the length of the flowering phase (PGS-6) was identical (59 days) in both sowing terms, indicating that while early vegetative development can be accelerated by later sowing, the reproductive duration remains largely stable. These findings highlight the plasticity of *E. ciliata* development in response to sowing date and environmental conditions, which is important for optimizing cultivation schedules for maximum biomass and essential oil yield.

The observed flowering pattern of *E. ciliata* is consistent with previous reports that classify it as a short-day

plant (Im et al., 2021). Under short-day conditions, flowering occurred 100% after nine weeks of sprouting, whereas extended photoperiods and night interruption effectively delayed flowering. This photoperiod sensitivity may explain the stability of the flowering duration observed in both sowing terms, as the plants likely initiated reproductive growth in response to naturally shortening daylengths during late summer. Comparisons with other *Elsholtzia* species also support the influence of environmental factors on reproductive timing. For instance, *E. splendens* displayed variation in the transition from vegetative to reproductive growth depending on altitude and cultivation method, with plants in plains regions flowering 22–29 days after vegetative onset and those in mid-mountain



**Figure 3** The height dynamics of the above-ground mass of *Elsholtzia ciliata* (Thunb.) Hyl. genotype 23–60 depends on sowing and irrigation dates at the 1<sup>st</sup> sowing date



**Figure 4** The height dynamics of the above-ground mass of *Elsholtzia ciliata* (Thunb.) Hyl. genotype 23–60 depends on sowing and irrigation dates at the 2<sup>nd</sup> sowing date

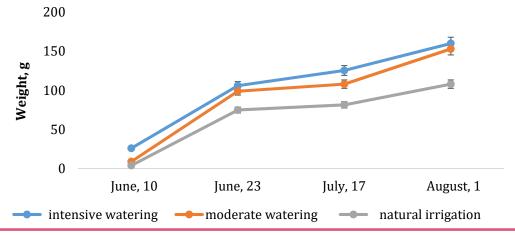
regions flowering after 26–35 days (Choi et al., 2024). These findings suggest that both photoperiod and local environmental conditions, such as temperature and altitude, play a key role in determining the onset and duration of flowering.

A notable distinction between sowing periods was that plants in the first sowing produced fewer inflorescences but exhibited greater foliage across all moisture regimes compared to the second sowing. Additionally, during the full flowering phase, plants from the second sowing under natural moisture conditions displayed reduced leaf turgor and occasional leaf yellowing, indicating possible water stress effects.

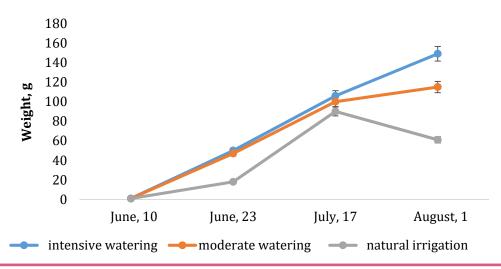
We studied the height of the investigated plants depending on the watering regime (Figures 3 and 4).

We found differences between plant height in the first and second sowing under intensive drop irrigation (IDI), moderate drop irrigation (MDI), and natural drop irrigation (NDI). In this case, plant growth accelerated in the tillering phase (PGS-2). In this phase, the height of the plants in the first sowing period varied from 7 to 17 cm, with a diameter of 12 to 25 cm. After two weeks, the height of the plants varied from 48 to 63 cm, with a diameter of 30 to 32 cm. The above-ground mass of the plants also increased, ranging from 75 to 106 g. In the mass flowering phase, the height of the plants varied from 85 to 90 cm, with a diameter of 46 to 57 cm. At the same time, the above-ground mass indicators ranged from 108 to 160 g per bush. Plants in the first sowing period had the highest indicators of bush habit and above-ground mass under intensive drip irrigation. In the process of plant development in the second sowing period, a gradual increase in plant height and diameter was also noted up to the mass flowering phase.

The above-ground biomass of individual plants varied between 4 and 26 g depending on sowing date (Figure 5 and 6). At the mass flowering stage, height ranged



**Figure 5** The accumulation of above-ground mass of *Elsholtzia ciliata* (Thunb.) Hyl. genotype 23-60 depends on sowing and irrigation dates at the 1<sup>st</sup> sowing date



**Figure 6** The accumulation of above-ground mass of *Elsholtzia ciliata* (Thunb.) Hyl. genotype 23–60 depends on sowing and irrigation dates at the 2<sup>nd</sup> sowing date

from 54 to 70 cm and diameter from 33 to 55 cm, with biomass of 61–149 g per plant (Figure 4B). For the second sowing, intensive drip irrigation again produced the highest growth and biomass, while natural moisture resulted in the lowest values. These findings indicate that water availability is a key factor in biomass accumulation, and intensive irrigation can enhance both vegetative growth and structural development. Furthermore, the earlier sowing promoted greater vegetative development, likely due to a longer period of favorable growth conditions, which is critical for optimizing cultivation schedules aimed at maximizing both biomass and essential oil yield in *E. ciliata*.

Plants of *E. ciliata* are herbs, subshrubs, or shrubs. Verticillasters in capitula or continuous or interrupted spikes; bracts oblong or flabellate to minute, thinner than calyx; spikes cylindric or second, frequently compact, occasionally in panicles. Calyx cylindric or campanulate, throat glabrous; 2 longer anterior or subequal teeth. The corolla is white, yellowish, or purplish, with two lips, hairy and glandular on the outside and hairy annulate or glabrous on the inside. The tube is funnel-shaped, straight or slightly curved, and slightly longer than the calyx. Nutlets are smooth to tuberculate, ovoid to oblong, glabrous, or sparingly hairy (Sahu and Sarankar, 2025).

Morphometric analysis of *E. ciliata* grown in Cherkasy Oblast demonstrated that plant architecture and organ dimensions were strongly influenced by both sowing date and irrigation regime (Table 2). Overall, plants under intensive drip irrigation exhibited the highest values for height, diameter, shoot number,

inflorescence dimensions, and leaf size, with the first sowing producing generally greater morphometric parameters than the second sowing. Exceptions were noted under intensive drip irrigation in the second sowing for specific traits, including first-order shoot number, inflorescence length of first-order shoots, inflorescence diameter of third-order shoots, and leaf length. Some traits, such as second-order inflorescence length and diameter, and leaf width, showed no significant differences between sowing dates under intensive irrigation.

Comparisons with published data indicate that the morphometric traits observed in Cherkasy are consistent with reported variability in E. ciliata. Korean plants ranged from 20-60 cm in height with leaf length of 3.0-9.0 cm and leaf width of 1.0-4.0 cm (Jeon and Hong, 2006). In Moldova, plants reached 50-80 cm in height and 40 cm in diameter at flowering (Colţun et al., 2021). Vietnamese plants exhibited lower height (28.34 cm), longer leaves (7.17 cm), and moderate leaf weight (4.22 g) (Luong et al., 2024). These comparisons suggest that environmental conditions, cultivation practices, and irrigation significantly modulate vegetative growth and morphometric traits in E. ciliata, emphasizing the importance of local adaptation for optimizing biomass and essential oil production.

The influence of irrigation on the green biomass of *Elsholtzia ciliata* was significant in both sowing periods. In the first sowing, plants and inflorescences exhibited the highest green mass under intensive drip irrigation (IDI) and the lowest under natural hydration (NH) (Figure 7).

Table 2 The morphometric parameters of *Elsholtzia ciliata* (Thunb.) Hyl. genotype 23–60 depends on sowing and irrigation dates

| Parameter                                                   | I                                     | First sowing date                       | e                                   | Second sowing date                    |                                         |                                     |  |
|-------------------------------------------------------------|---------------------------------------|-----------------------------------------|-------------------------------------|---------------------------------------|-----------------------------------------|-------------------------------------|--|
|                                                             | intensive drip<br>irrigation<br>(±SD) | moderate<br>drip<br>irrigation<br>(±SD) | natural drip<br>irrigation<br>(±SD) | intensive drip<br>irrigation<br>(±SD) | moderate<br>drip<br>irrigation<br>(±SD) | natural drip<br>irrigation<br>(±SD) |  |
| Plant height, cm                                            | 90.03 ±2.18                           | 90.55 ±3.47                             | 85.15 ±4.87                         | 68.44 ±2.45                           | 62.13 ±3.16                             | 57.16 ±2.78                         |  |
| Plant diameter, cm                                          | 57.11 ±3.27                           | 50.18 ±2.19                             | 46.13 ±1.21                         | 42.01 ±2.76                           | 42.98 ±1.76                             | 40.98 ±0.56                         |  |
| Number of first-order shoots, pcs                           | 22.10 ±0.97                           | 20.11 ±0.34                             | 18.16 ±0.45                         | 24.12 ±0.67                           | 18.09 ±1.13                             | 16.08 ±0.67                         |  |
| Number of second-order shoots, pcs                          | 110.25 ±6.54                          | 90.55 ±4.78                             | 60.89 ±4.65                         | 102.17 ±6.72                          | 86.09 ±4.56                             | 56.09 ±2.78                         |  |
| Number of third-order shoots, pcs                           | 200.18 ±8.45                          | 180.34 ±12.89                           | 130.87 ±7.98                        | 187.56 ±13.09                         | 172.12 ±5.57                            | 122.09 ±8.45                        |  |
| Number of inflorescences per plant, pcs                     | 296.11 ±15.18                         | 282.12 ±9.98                            | 209.11 ±10.45                       | 290.56 ±13.19                         | 258.67 ±20.14                           | 201.32 ±16.87                       |  |
| Inflorescence length of first-order shoot, cm               | 6.61±0.45                             | 6.04 ±0.45                              | 5.04 ±0.23                          | 7.21 ±0.45                            | 6.94 ±0.23                              | 4.84 ±0.56                          |  |
| Inflorescence diameter of first-order shoot, pcs            | 1.63 ±0.04                            | 1.65 ±0.11                              | 1.47 ±0.05                          | 1.6 ±0.05                             | 1.5 ±0.07                               | 1.2 ±0.02                           |  |
| Inflorescence length of second-order shoot, cm              | 5.06 ±0.23                            | 4.54 ±0.17                              | 2.04 ±0.11                          | 5.06 ±0.16                            | 4.37 ±0.21                              | 4.03 ±0.11                          |  |
| Inflorescence diameter of second-order shoot, pcs           | 1.27 ±0.03                            | 1.09 ±0.03                              | 0.65 ±0.13                          | 1.27 ±0.06                            | 1.07 ±0.09                              | 0.55 ±0.08                          |  |
| Inflorescence length of 3 <sup>rd</sup> -order shoot, cm    | 3.74 ±0.18                            | 3.54 ±0.13                              | 3.38 ±0.14                          | 3.68 ±0.14                            | 3.49 ±0.09                              | 3.03 ±0.07                          |  |
| Inflorescence diameter of 3 <sup>rd</sup> -order shoot, pcs | 0.66 ±0.02                            | 0.62 ±0.06                              | 0.59 ±0.07                          | 0.83 ±0.04                            | 0.88 ±0.02                              | 0.59 ±0.07                          |  |
| Leaf length, cm                                             | 5.54 ±0.32                            | 5.55 ±0.11                              | 3.57 ±0.24                          | 5.57 ±0.21                            | 5.56 ±0.13                              | 3.57 ±0.12                          |  |
| Leaf width, cm                                              | 2.56 ±0.14                            | 2.75 ±0.16                              | 1.79 ±0.03                          | 2.56 ±0.05                            | 2.47 ±0.03                              | 1.68 ±0.09                          |  |

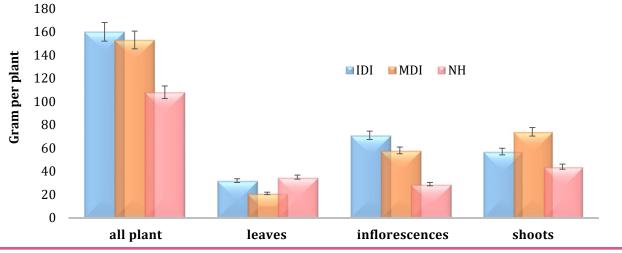


Figure 7 The yield of green mass of *Elsholtzia ciliata* (Thunb.) Hyl. genotype 23–60 on first sowing, depending on irrigation dates
IDI – intensive drip irrigation; MDI – moderate drip irrigation; NH – natural hydration

Interestingly, leaf and shoot mass reached maximal values under moderate and natural watering, suggesting differential allocation of assimilates between organs depending on water availability. Overall, the green mass yield ranged from 108 to 160 g per plant, with leaves contributing 21–35 g, inflorescences 29–71 g, and shoots 44–74 g. These results are higher than those reported by Luong et al. (2024), who observed 32.36 g per plant after 8 weeks, likely reflecting differences in genotype, environmental conditions, and cultivation practices.

Similarly, during the second sowing, intensive drip irrigation promoted the highest green mass of plants, inflorescences, and shoots, while natural hydration resulted in the lowest values (Figure 8). Leaf yield was maximal under moderate drip irrigation. Total green mass per plant ranged from 61 to 149 g, leaf mass from 19 to 30 g, inflorescences from 14 to 55 g, and shoots from 28 to 66 g. These findings confirm that water regime is a key factor influencing biomass partitioning in *E. ciliata*, consistent with observations that irrigation positively affects growth and productivity of aromatic and medicinal plants (Khalid, 2006; Askary et al., 2018).

Essential oil is one of the most valuable raw materials of *E. ciliata*, with documented hypotensive (Martišiene et al., 2023), anti-inflammatory (Nguyen et al., 2021), insecticidal (Song et al., 2024), anticancer, and antimicrobial (Nguyen et al., 2025) activities. The main components include (Z)- $\beta$ -Farnesene (22.72%), neral (15.66%), geranial (15.62%), and  $\beta$ -ocimene (13.30%) (Khang et al., 2023; Tran et al., 2024).

The content of essential oil depends on multiple factors, including growth conditions, climate, plant density, sowing date, and irrigation regime (Zawiślak and Nurzyńska-Wierdak, 2017; Morteza et al., 2009). Studies indicate that late sowing and low planting density can enhance essential oil yield, while water availability modulates both quantity and quality (Khalid, 2006; Askary et al., 2018).

In our experiments, essential oil content increased throughout the growing season, reaching a maximum during mass flowering. In the first sowing, the mass fraction of essential oil ranged from 0.45 to 0.82%, whereas in the second sowing, values ranged from 0.57 to 0.59% (Figure 9). Notably, no significant differences were observed among irrigation treatments in the second sowing, suggesting that sowing date may have a stabilizing effect on reproductive secondary metabolite accumulation.

Comparison with published data shows variation in essential oil content across regions. In Moldova, *E. ciliata* achieved 1.5–2.5% essential oil, with the highest concentration in inflorescences during full flowering (Colţun et al., 2021). Khang et al. (2023) reported 0.82% essential oil in leaves, comparable to our IDI and MDI results for the first sowing. Vietnamese plants showed 0.93% in control and 1.41% after gibberellic acid treatment (Luong et al., 2024). These differences likely reflect genotype, environmental conditions, plant development stage, and cultivation practices. Overall, the data indicate that intensive irrigation and proper sowing date can optimize both green biomass and essential oil yield in *E. ciliata*,

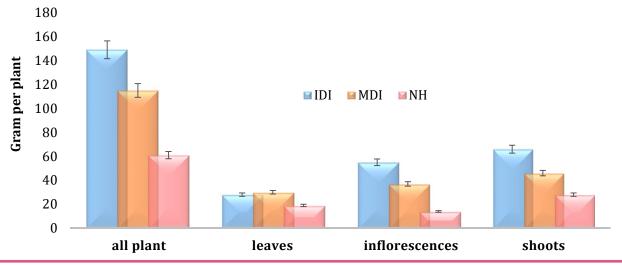
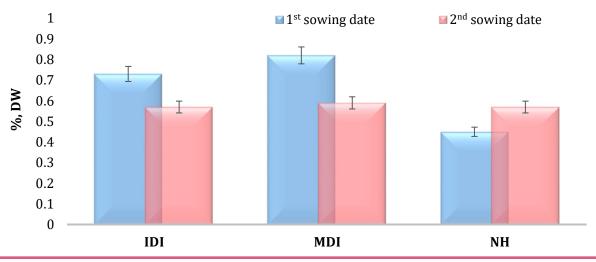


Figure 8 The yield of green mass of *Elsholtzia ciliata* (Thunb.) Hyl. genotype 23–60 on the second sowing, depending on irrigation dates
IDI – intensive drip irrigation; MDI – moderate drip irrigation; NH – natural hydration



**Figure 9** The mass fraction of essential oil of *Elsholtzia ciliata* (Thunb.) Hyl. genotype 23–60 depending on sowing and irrigation dates

DW - dry weight; IDI - intensive drip irrigation; MDI - moderate drip irrigation; NH - natural hydration

confirming its potential as a valuable aromatic and medicinal plant.

#### **Conclusions**

The study demonstrated that sowing date and irrigation regime significantly influence the growth, biomass accumulation, and essential oil content of Elsholtzia ciliata in the Central Forest-Steppe region. Plants sown in the first period under intensive drip irrigation exhibited the highest height, diameter, and above-ground biomass yield, whereas those from the second sowing under natural hydration produced the lowest biomass. The mass fraction of essential oil peaked in plants of the first sowing under moderate drip irrigation, highlighting the potential of optimized irrigation to enhance phytochemical quality. These findings provide important guidance for cultivation practices aimed at maximizing biomass and essential oil yield. Furthermore, the results can inform breeding programs focused on selecting genotypes with superior productivity and phytochemical content under diverse environmental and agronomic conditions. Future studies could investigate interactions between sowing dates, irrigation intensity, and other environmental factors, such as light and soil fertility, to refine cultivation strategies for sustainable production of high-value plant raw materials.

# Conflict of interest

The authors declare no conflict of interest.

#### **Ethical Statement**

This article does not contain any studies that would require an ethical statement.

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