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Optimization of Plant Density and Harvest Time to Maximize Volatile Oil Accumulation in Two Aromatic Plants

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Abstract: Volatile oil, mainly extracted from aromatic plants, is widely used in medical, cosmetics, and other industries for its disinfection, sterilization, antioxidant, and other effects. The yield of volatile oil was influenced by plant density and harvest time. Here, two common aromatic plants, *Perilla frutescens* and *Ysphania ambrosioide*, were taken as examples to investigate the effects of plant density and harvest time on plant yield-related traits, content, and yield of volatile oil, and the oil yield was determined by steam distillation. Results showed that plant density and harvest time significantly affected the yield and quality of the two plants ($p < 0.05$). Increasing plant density significantly increased the dry yield and leaf dry yield of *P. frutescens*, while the dry yield and volatile oil content and yield of *Y. ambrosioides* significantly decreased, and the maximum value exhibited was at the plant density of 80 cm × 80 cm. In *P. frutescens*, the maximum volatile oil content was obtained at the plant density of 60 cm × 60 cm, and the maximum volatile oil yield was obtained at the plant density of 30 cm × 30 cm. The yield and volatile oil yield of these two plants increased first and then decreased with the extension of harvest time. The maximum of *P. frutescens* was in mid-September, and that of *Y. ambrosioides* was in late October to early November. Our study provides new data for improving plant volatile oil yield and commercial value by optimizing planting density and harvest time and highlights the importance of optimizing planting management mode.

Keywords: volatile oil; plant density; harvest time; volatile oil yield; *Perilla frutescens*; *Ysphania ambrosioides*

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1. Introduction

Volatile oil (VO), also known as essential oil, is an aromatic oily liquid and a mixture of complex compounds extracted from the flowers, leaves, roots, stems, fruits, seeds, and other parts of aromatic plants [1]. Most VOs have an aromatic odor and are of high medical value, with important roles in anti-inflammatory, anti-allergic, anti-cancer, and antioxidant medical treatments. In addition, there is a long history of utilizing volatile oils in the treatment of emotional disorders around the world [2–4].

Perilla frutescens (L.) Britt. is an annual herb commonly known as perilla, kaennip, zisu, and shiso and is known for its essential oils. *P. frutescens* has strong adaptability, is native to China, and was later widely naturalized in other Asian countries. Currently, 193 compounds have been isolated and identified from *Perilla frutescens*, including flavonoids, phenolic acids, triterpenoids, and so on. In addition, the volatile oil extracted from it is rich in components such as monoterpenoid perillaldehyde and bicyclic sesquiterpenes β-caryophyllene and has a unique flavor [5–7]. It has also been cultivated on a large scale in European countries [8]. The complex volatile compounds in the VO of *P. frutescens* possess various functions; they contain anti-allergy, hypolipidemic, anti-aging, cardiovascular disease prevention, and other functions [9]. It can be widely used in food, cosmetics, medical, and other industries and has significant economic value [10,11]. *Ysphania ambrosioides*

(L.) Mosyakin et Clemants is an annual or perennial herb with a strong fragrance [12]. *Y. ambrosioides*, native to tropical America, is now widely distributed in tropical and temperate regions [13]. The whole plant of *Y. ambrosioides* contains volatile oil, which is the most abundant in the fruits [14]. It is also an edible and medicinal plant with important commercial value. The VO of *Y. ambrosioides* has a variety of pharmacological activities such as anti-parasitic, antibacterial, dehumidification, and so on [1,15], and is often used as a condiment in some countries because of its pungent and distinctive flavor [16]. Previous studies on *Y. ambrosioides* have mainly focused on the composition of VO and its medicinal value [17,18]. However, there is still no research on the planting density and harvesting time of *Y. ambrosioides.* Along with an increase in the demand for VO and labor costs and the tighter land resources, an intensive and standardized planting mode is inevitable for increasing yield and quality. Hence, adopting more efficient and resource-efficient cultivation patterns is essential to producing high-quality crops on a large scale and satisfy industrial demand.

This study aimed to evaluate the effects of plant density and harvest time on VO content and yield. Here, we focused on the plant yield, VO content, and VO yield under different plant densities and harvest times, taking the valuable crops *Perilla frutescens* and *Ysphania ambrosioides* cultivated worldwide as examples. The results could provide a reliable scientific basis for efficient and standardized cultivation of *P. frutescens* and *Y. ambrosioides* in production practice and further offer new data for improving VO's high yield and efficiency by optimizing plant density and harvest time.

2. Materials and Methods

2.1. Plant Materials and Site Description

Perilla frutescens (*P. frutescens*) and *Ysphania ambrosioides* (*Y. ambrosioides*) used in this study were sown in mid-March 2018 in the Shaanxi Tasly Plant Pharmaceutical Co., Ltd. (Shangluo, China). (Engineer HongGuang Zhao identified all plant samples). The seeds of *Ysphania ambrosioides* and *Perilla frutescens* are varieties that have been selected and saved by Shaanxi Tasly Plant Pharmaceutical Co., Ltd. (Shangluo, China). The study area was located in Shangzhou, Shaanxi Province, China (38°87′ N, 109°93′ E, and 708 m elevation). It is characterized by a warm temperate semi-humid monsoon climate, with an average annual temperature of 12.8 °C and mean annual precipitation of 700 mm (mainly falling between July and September). The two crops were grown in greenhouses, where temperature and humidity were controlled at daytime temperatures of 23–28 °C, nighttime temperatures of about 18 °C, and relative air humidity of about 75%.

The soil pH for planting *P. frutescens* and *Y. ambrosioides* was 5.31, and seeding was performed by direct seeding. For sowing, the substrate soil material was vermiculite, grass charcoal, and perlite, with a mixing ratio of 5:10:2. Fill the 72-hole disk tray with substrate soil, lightly press the substrate soil inside the tray, and sow 1 to 2 seeds per hole. Cover with vermiculite after sowing and water in time. When the plants in the 72-hole trays reach a height of about 20 cm and have 5–8 leaves in about 20 days, they can be planted. Dig a planting hole, with the depth of the hole at about 7 cm, plant the planting seedlings into the hole, and compact it. Water well after planting to facilitate the recovery of the planted seedlings. The ratio of nitrogen, phosphorus, and potassium fertilizers was 1.5:1.0:2.0. Fertilizer was applied in the middle of every 4 plants, the hole depth was about 10 cm, 0.05 kg of compound fertilizer was applied to the hole, the soil was covered and compacted, and the soil was watered in a timely manner after the fertilizer was applied.

2.2. Plant Density of *P. frutescens* and *Y. ambrosioides*

This experiment adopted a completely randomized block design with five density gradients, and each experiment was repeated three times. The plant density of *P. frutescens* was 30 cm × 30 cm, 40 cm × 40 cm, 50 cm × 50 cm, 60 cm × 60 cm, 70 cm × 70 cm (hereafter referred to as PD1, PD2, PD3, PD4, and PD5, respectively) and the plant density of *Y. ambrosioides* was 50 cm × 50 cm, 60 cm × 60 cm, 70 cm × 70 cm, 80 cm × 80 cm, 90 cm × 90 cm (hereafter referred to as YD1, YD2, YD3, YD4, and YD5, respectively). The fresh weights of *P.* frutescens and *Y. ambrosioides* were measured in a 30 m^2 sampling area per replication. Traits were evaluated for five individual plants per replication, including plant dry weight, fresh weight, leaf dry weight, leaf fresh weight, fresh seed weight, and dried seed weight. The VO content and VO yield were calculated as indexes to screen out the suitable plant density of *P.* frutescens and *Y. ambrosioides*. The calculation formula of VO yield and essential oil content was calculated in v/w (%) as follows:

$$\text{VO yield}\ (\text{kg ha}^{-1}) = \frac{10{,}000}{\text{sampling area}} \times \text{sampling area leaf (seed) dry weight} \times \text{VO content} \tag{1}$$

$$\text{VO content of } P.\,frutescens\ (‰) = \frac{\text{Extracted volatile oil volume (mL)}}{\text{dry leaves (g)}} \times 1000‰ \tag{2}$$

$$\text{VO content of } Y.\,ambrosioides\ (\%) = \frac{\text{Extracted volatile oil volume (mL)}}{\text{dry seeds (g)}} \times 100\% \tag{3}$$

2.3. Harvest Time of P. frutescens and Y. ambrosioides

P. frutescens was harvested about every 15 days from late August to late October, and *Y. ambrosioides* was harvested about 20 days from early September to early December (the harvest interval was adjusted according to the weather), and each experiment was repeated three times. The fresh weights of *P.* frutescens and *Y. ambrosioides* were measured in a 30 m^2 sampling area per replication. Traits were evaluated for five individual plants per replication, including plant dry weight, fresh weight, leaf dry weight, leaf fresh weight, fresh seed weight, and dried seed weight. The VO content and VO yield were calculated as indexes to screen out the suitable harvest time of *P. frutescens* and *Y. ambrosioides*.

2.4. Material Drying Methods and Extraction of Volatile Oils

Fresh *P. frutescens* was subjected to stem and leaf separation, and seeds were harvested from *Y. ambrosioides*. The leaves of *P. frutescens* were dried by natural drying for 3–4 d until the quality of the leaves no longer changed, and the seeds of *Y. ambrosioides* were picked and washed and dried by natural shade-drying until the quality of the seeds no longer changed. In this experiment, the seeds were dried immediately after harvesting to minimize the influence of external factors on the essential oil content and composition.

The VO of *P. frutescens* and *Y. ambrosioides* were extracted by steam distillation. Steam distillation is a standard extraction method of VO, which can effectively improve the recovery rate of VO (Valderrama, 2018). Moreover, 200.00 g dry leaves of *P. frutescens* and 100.00 g dry seeds of *Y. ambrosioides* were accurately weighed and placed in two 5000 mL round-bottomed flasks. Furthermore, 3000 mL and 1500 mL of distilled water were put into the flask with *P. frutescens* and *Y. ambrosioides*. Then, we added several glass beads, shook them to the mix, and connected the volatile oil detector and reflux condenser. We added water from the upper end of the condenser tube to make it full of the scale part of the volatile oil detector and overflow into the flask. The set was placed in an electric heating mantle and kept slightly boiling for 3 h until the oil no longer increased. After cooling to room temperature and the oil–water was completely separated, the piston was turned on to slowly release the water layer until the upper end reached 5 mm on the 0 line of the scale; we let it stand for more than 1 h. We continued to turn on the piston until the upper end of the oil layer was flush with the scale 0 line and recorded the volatile oil volume. The VO content from leaves of *P. frutescens* and dry seeds of *Y. ambrosioides* were calculated as follows:

2.5. Data Analysis

Data were represented as mean ± standard error (SE). One-way ANOVA was applied to determine the impacts on volatile oil content, volatile oil yield, and other parameters induced by plant density and harvest time, and Duncan's test was performed for multiple comparisons ($p < 0.05$) using SPSS Statistics 22.0 (IBM). Graphs were plotted using Origin 2021.

3. Results

3.1. Effects of Different Plant Density and Harvest Time on the Yield of *P. frutescens* and *Y. ambrosioides*

The effects of plant density and harvest time on yield-related traits of *P. frutescens* and *Y. ambrosioides* are shown in Table 1 and Figure 1. Specifically, increasing plant density significantly increased the dry yield and leaf dry yield of *P. frutescens* ($p < 0.05$, Figure 1a). Still, the fresh yield and leaf-to-stem ratio showed no significant difference (Table 1). The plant density also had no significant effect on fresh yield and percentage of seed of *Y. ambrosioides*, while the dry leaves yield and seed dry yield were significantly decreased with an increase in plant density ($p < 0.05$, Figure 1b). In addition, the harvest time significantly affected the yield-related traits of *P. frutescens*, increasing first and then decreasing, and the maximum dry weight and leaf dry yield existed on 11 September (Figure 1c). Similar to *P. frutescens*, the yield traits of *Y. ambrosioides* showed a single peak curve with harvest time, but the percentage of seed did not change significantly (Figure 1d).

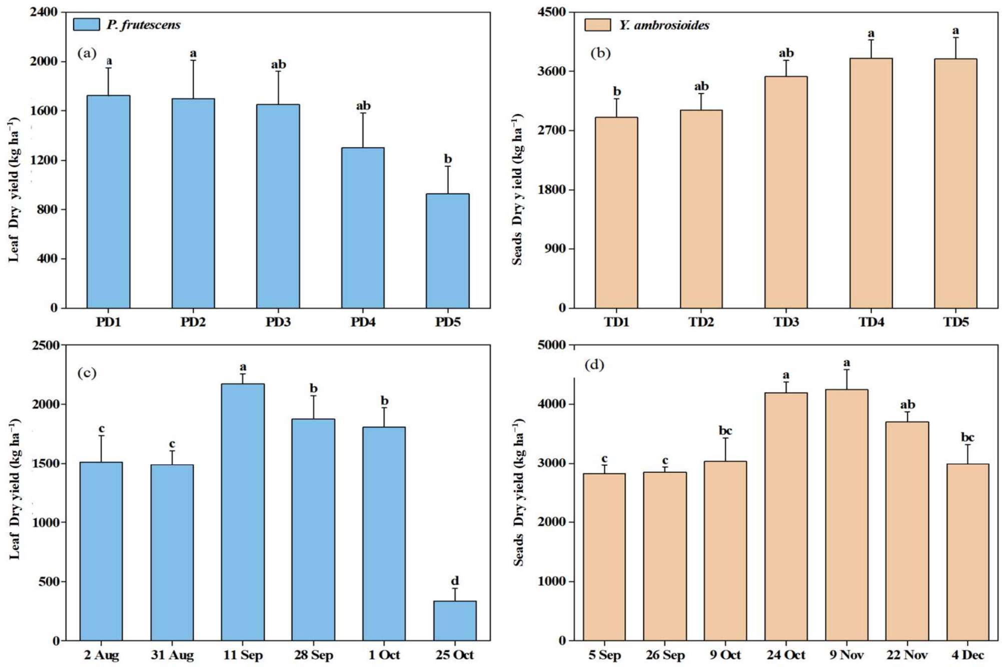

Figure 1. Leaf dry yield of *P. frutescens* and seed dry yield of *Y. ambrosioides* under different plant density and harvest time treatment. (**a**,**b**) Dry leaf yield as well as dry seed yield of *P. frutescens* and *Y. ambrosioides* at PD1-PD5 and TD1-TD5 planting densities. (**c**,**d**) Effect of different harvest time on dry leaf as well as dry seed yield of *P. frutescens* and *Y. ambrosioides*. Error bars denote standard error. Different letters indicate significant differences at $p < 0.05$ in different treatments.

Table 1. The effects of harvest time on yield-related traits of *P. frutescens* and *Y. ambrosioides*.

Plant Species	Harvest Time	Fresh Leaves (Seeds) Yield (kg ha^{-1})	Dry Leaves (Seeds) Yield (kg ha^{-1})	Stages of Plant Growth
P. frutescens	2 Aug	661.5 ± 93.18c	237.02 ± 34.49b	Nutrient growth period
	31 Aug	575.90 ± 50.32c	252.07 ± 20.94b	
	11 Sep	865.16 ± 49.40bc	388.65 ± 19.98a	Flowering period
	28 Sep	1228.86 ± 68.78a	358.35 ± 27.49a	Prime bloom period
	1 Oct	1341.97 ± 110.46a	405.65 ± 36.41a	Final flowering period
	25 Oct	1077.20 ± 102.59ab	378.11 ± 27.52a	Seed maturation period

Table 1. *Cont.*

Plant Species	Harvest Time	Fresh Leaves (Seeds) Yield (kg ha^{-1})	Dry Leaves (Seeds) Yield (kg ha^{-1})	Stages of Plant Growth
Y. ambrosioides	5 Sep	1287.67 ± 57.49b	421.20 ± 19.46b	
	26 Sep	1317.96 ± 44.50b	430.44 ± 16.32b	
	9 Oct	1253.90 ± 109.18b	445.94 ± 56.52b	
	24 Oct	1997.30 ± 145.62a	582.25 ± 29.362a	Seed maturation period
	9 Nov	1969.19 ± 163.04a	591.89 ± 58.10a	
	22 Nov	1557.14 ± 113.64b	518.80 ± 38.10ab	
	4 Dec	828.36 ± 59.26c	437.20 ± 40.07b	

Values are the means ± standard error, Different letters indicate significant differences at $p < 0.05$ in different treatments.

3.2. *Effects of Different Plant Density and Harvest Time on VO Content of P. frutescens and Y. ambrosioides*

Plant density and harvest time had significant effects on the VO content of *P. frutescens* and *Y. ambrosioides* ($p < 0.05$, Figure 2). For *P. frutescens*, the plant density of 60 cm × 60 cm (PD4) exhibited the maximum VO content (10.17 ‰), while the minimum VO content (7.94 ‰) at the plant density of 70 cm × 70 cm (PD5). There was no significant difference among the other three planting densities (Figure 2a). Furthermore, the VO content of *Y. ambrosioides* decreased significantly with increasing plant density ($p < 0.05$, Figure 2b), and the maximum VO content (1.74%) was obtained at the plant density of 80 cm × 80 cm (YD4). VO content refers to the proportion of essential oils obtained from a plant and can reflect the plant's ability to synthesize its own essential oils under the influence of different factors. Therefore, with VO content as the index, the suitable plant density of *P. frutescens* and *Y. ambrosioides* should be 60 cm × 60 cm and 80 cm × 80 cm, respectively.

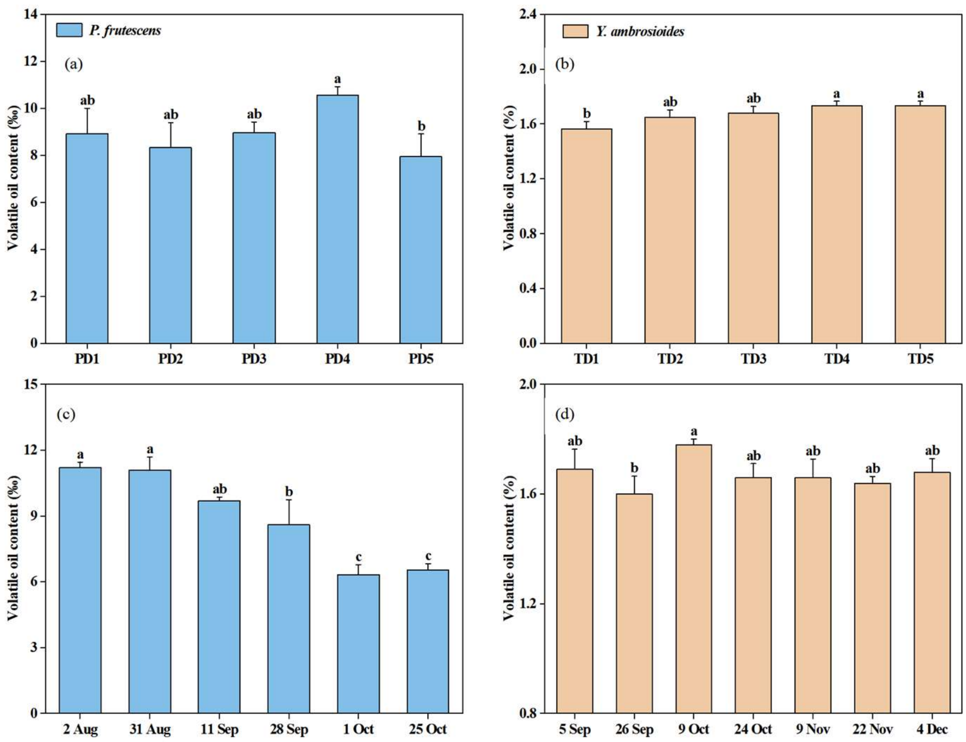

Figure 2. VO content of *P. frutescens* and *Y. ambrosioides* under different plant densities and harvest time treatments. (**a**,**b**) VO content of *P. frutescens* and *Y. ambrosioides* at TD1-TD5 and PD1-PD5 planting densities. (**c**,**d**) VO content of *P. frutescens* and *Y. ambrosioides* obtained at different harvest periods. Error bars denote standard error. Different letters indicate significant differences at $p < 0.05$ in different treatments.

Tracing the data revealed that the VO content of *P. frutescens* significantly decreased with the extension of harvest time ($p < 0.05$, Figure 2c). The highest VO content was 11.20‰ in the first harvest time (August 20th), and slightly decreased to 11.09‰ in the second harvest time (13 August), since then significantly reduced with a drop of more than 1.1‰. Thus, taking the VO content as the index, the suitable harvest time of leaves of *P. frutescens* was late August. However, with the extension of the harvest time, the VO content of dry seeds of *Y. ambrosioides* had no obvious change rule; that is, the harvest time had little effect on the oil production rate of *Y. ambrosioides* (Figure 2d).

3.3. Effects of Different Plant Density and Harvest Time on VO Yield of P. frutescens and Y. ambrosioides

The VO yield of *P. frutescens* and *Y. ambrosioides* was significantly influenced by plant density and harvest time ($p < 0.05$, Figure 3). In the range of planting density from PD1 (30 cm × 30 cm) to PD4 (60 cm × 60 cm), the VO yield of *P. frutescens* reached more than 13.86 kg ha^{-1} (Figure 3a). Among these, the highest value (15.89 kg ha^{-1}) was found at the plant density of PD1 (30 cm × 30 cm), while only 7.21 kg ha^{-1} under the plant density of PD5 (70 cm × 70 cm). The VO yield of *Y. ambrosioides* significantly increased first and remained stable with the decrease in planting density ($p < 0.05$, Figure 3b). The highest VO yield was 66.02 kg ha^{-1} at the planting density of YD4 (80 cm × 80 cm), and the lowest VO yield was 45.16 kg ha^{-1} under the planting density of YD1 (50 cm × 50 cm). Therefore, regarding the highest VO yield as the index, the suitable plant density of *P. frutescens* and *Y. ambrosioides* could be 30 cm × 30 cm and 80 cm × 80 cm, respectively.

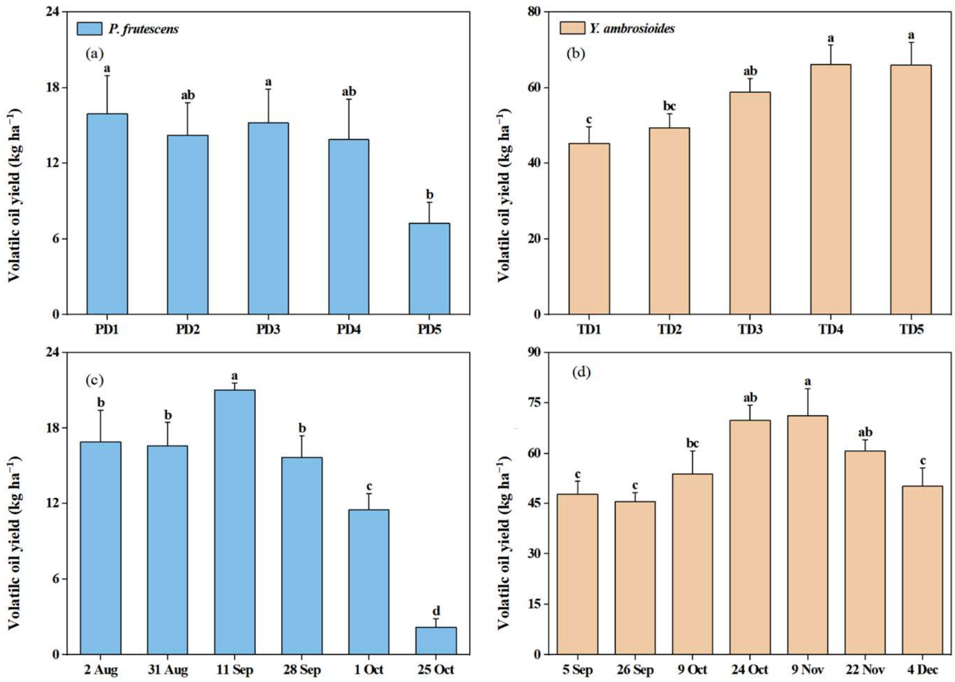

Figure 3. VO yield of *P. frutescens* and *Y. ambrosioides* under different plant density and harvest time treatment. (**a**,**b**) VO yields of *P. frutescens* and *Y. ambrosioides* at PD1-PD5 and TD1-TD5 planting densities. (**c**,**d**) VO yields of *P. frutescens* and *Y. ambrosioides* at different harvest time. Error bars denote standard error. Different letters indicate significant differences at $p < 0.05$ in different treatments.

Meanwhile, the VO yield of *P. frutescens* increased first and then decreased with the harvest time, reaching a peak (21.03 kg ha^{-1}) on 11 September (Figure 3c). The variation tendency of the VO yield of *Y. ambrosioides* was similar to *P. frutescens,* which increased first and then decreased. In detail, the rising stage was from September to the end of October, the peak value was 18.01 kg ha^{-1} on November 9th, and the decreasing stage was from early November to December (Figure 3d). Therefore, taking the VO yield as the index, the suitable harvest time of *P. frutescens* and *Y. ambrosioides* was about September 11th, the end of October, and early November, respectively.

4. Discussion

The yield of aromatic herbs and VO accumulation are influenced by various factors, including plant genotype, climatic conditions, growth stage, and cultivation and management patterns [19]. Plant density and harvest time are two of the essential management factors that are closely related to plant growth and nutrition [20,21]. Planting density mainly affects water, nutrient, and light competition during plant growth and, finally, affects crop yield [22]. In this study, increasing plant density significantly increased the dry yield and leaf dry yield of *P. frutescens*, while the dry yield and seed dry yield of *Y. ambrosioides* significantly decreased ($p < 0.05$, Table 1, Figure 1). This is mainly related to plant-growing properties. *P. frutescens* has strong adaptability, favors warm and humid climates, and can grow in the shade [23]. Although the decrease in plant space reduced the photosynthesis of individual plants, the population quantity advantage made up for the insufficient biomass accumulation of particular plants, which showed an increase in yield. Nurzynska-Wierdak and Zawislak (2014) also showed that shoot dry weight and leaf dry weight per unit area increased as plant density increased. Some studies have shown that high-density planting can improve the shading rate, the growth rate of crops, and the biomass of crops, which can improve the total yield of crops [24]. However, *Y. ambrosioides* grows in the sun, and the larger plant density can reduce leaves and the number of branches due to shading and lower light interception, which affects plant growth. This is consistent with previous research, which reported that with increasing plant density, the number of branches decreased, and dry weight per plant decreased above ground [25,26]. Moreover, with the increase in row spacing, competition for natural resources such as light, water, and nutrition also decreased. Thus, the fresh weight and dry weight of herbs increased [20,22].

In general, the quality of the aromatic plants is evaluated by VO content and VO yield [27,28]. The biosynthesis of volatile oil may differ depending on the plant under different planting densities. The lower the planting density, the higher the growth and the VO content per plant, while increasing plant density increases crop yield and VO yield per unit area [29]. Our results showed that although the weight of *P. frutescens* per plant decreased significantly with increasing planting density, the VO content also showed a decreasing trend. However, the quantity of harvested plants increased, and dry yield per unit area increased, ultimately leading to a significant increase in VO yield ($p < 0.05$, Figure 2c). However, when the planting density exceeds a certain limit, the adequate nutrients of individual plants decrease correspondingly. Ventilation rates and photosynthesis are also reduced, making them more susceptible to pests and diseases, which reduces the width and weight of the leaves of *P. frutescens* [25]. The VO content of *P. frutescens* reached the maximum at PD4, and 60 cm $\times$ 60 cm was the optimum plant density when planting for the volatile oil production industry. And the maximum VO yield was found at PD1; 30 cm $\times$ 30 cm was the suitable planting density when dry leaves were used as spices. As for *Y. ambrosioides*, the maximum VO content and VO yield were all obtained at the planting density of YD4 (80 cm $\times$ 80 cm), and the higher the planting density, the lower the VO content and VO yield. Notably, the percentage of seeds also declined (Table 1). These observations confirmed that with the increase in plant density, the availability of ecological resources such as light, water, and nutrients was limited. Plants began to compete with each other for these resources, resulting in slow plant development and limited seed growth [25]. The accretion population quantity is insufficient to make up for the inadequacy of individual plants and thus reduced oil yield [30]. It also further indicated that different plants have different carrying capacities to resource competition caused by dense planting. This means that VO accumulation can be enhanced by optimizing the planting density of the plants.

The stage of growth of the aromatic plant has a similarly significant influence on the final VO yield and VO content [31]. Previous studies found that more mature parsley had higher VO content [32]. The VO content of lavender at the end of the flowering phase was higher than the bud and full flowering phases [21]. The determination of VO content in the leaves of *Rhododendron tomentosum* at different growth periods by Zhao et al. showed that the volatile oil content in the leaves peaked during the nutritive growth phase and would decline once the reproductive phase was entered. Shao et al. showed that the VO content of each organ was the lowest during the rapid growth stage of *P. frutescens* [33]. By the nutrient growth stage of *P. frutescens*, the VO content in each part was significantly higher than that in other growth stages, and the highest VO content was extracted from the leaves. Further, after the flowering stage, there was a decrease in the VO content in the leaves of *P. frutescens*, which may be due to the shift of the plant's primary nutrients to reproduction. Therefore, the relationship between VO content and yield needs to be considered together to further determine the optimal timing of harvest. Furthermore, Wei et al. showed that among the different growth stages of *P. frutescens*, the highest VO content was found in the leaves at the end of the nutrient stage as well as in the middle of the pre-flowering stage [34]. And when the growth stage shifted to seed maturity, the VO content would decrease sharply. This is consistent with our findings. In addition, plant antagonisms are a critical factor affecting the yield and composition of VO [35,36]. Therefore, it will be necessary to clarify the relationship between VO yield and quality in more ways. Quality will be necessary.

In the present research, the harvest time significantly affected some yield-related traits of *P. frutescens* and *Y. ambrosioides* ($p < 0.05$), showing a trend of increasing first and then decreasing [37] (Figure 1). The highest VO content of *P. frutescens* was found in the first harvest time and greatly reduced after September 28th (Figure 2c). In other words, the maximum VO content reached the bud flowering stage, which was higher than the full and end of the flowering stage, but the leaf dry yield and dry yield were highest at the full flowering stage, so the VO yield reached the maximum. This is similar to the VO accumulation of thyme; the maximum VO content and thymol yield were obtained at the beginning of the blooming stage [38]. Moreover, the maximum oregano yield was exhibited at the entire flowering stage, and the VO yield reached the maximum [39]. The subsequent decline in VO content and yield can be explained by precipitation, which negatively correlates with VO accumulation [21].

In this experiment, the rainfall before full flowering was less than that at the end of flowering. Hence, the suitable harvest time was mid-September, when both VO content and yield reached the best standards. However, the VO content of the dry seeds of *Y. ambrosioides* had no obvious change rule, and the VO yield increased first and then decreased (Figures 2d and 3d). The VO yield was the highest from late October to early November, which further confirmed that appropriate harvest time increased the VO accumulation. Seasonal variation was observed in the berry oils of Juniperus, and the VO yield increased with berry ripening [40]. Overall, a suitable harvest time should be recommended based on the use of aromatic plants, planting conditions, and plant properties.

5. Conclusions

In this study, we learned that plant density and harvest time had impacts on the yield-related traits, VO content, and VO yield of *P. frutescens* and *Y. ambrosioides*. Increasing plant density increased the dry yield and leaf dry yield of *P. frutescens*. The VO content reached the maximum at the plant density of 60 cm × 60 cm, which can reduce the production cost for VO production. The maximum VO yield was at 30 cm × 30 cm, which can be encouraging to farmers, leading to an improvement in the utilization rate of land mobilization, and it can be suitable for dry leaf spices. As for *Y. ambrosioides*, the plant yield, VO content, and yield decreased with the increase in plant density. This was mainly due to the different growth characteristics; different plants have different carrying capacities to resource competition caused by dense planting. These two plants' yield and VO yield increased first and then decreased with the extension of harvest time. The maximum of *P. frutescens* was in the middle of September, and that of *Y. ambrosioides* was in late October to early November. Our study provided a reference for suitable plant density and harvest time of *P. frutescens* and *Y. ambrosioide*. It highlighted that the yield and quality of aromatic plants can be improved by optimizing plant density and harvest time. However, the effects of plant density and harvest time on VO composition need further study.

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