


## Article

# Pollination and Seed Production of *Lavandula angustifolia* Mill. (Lamiaceae)

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**Abstract:** *Lavandula angustifolia* Mill., lavender, is an aromatic plant in the Lamiaceae family. Lavender is an important economic plant that is cultivated throughout the world. Previous studies have shown that the primary pollinators of lavender in Europe and North Africa are bee species, specifically *Bombus* spp. However, similar studies have not been previously performed in North America. The current study, on cultivated population lavender (grown from seed) in Utah (USA), found a diverse community of bees visiting lavender over a 4-week sequential and successive blooming period. The observed and identified bees were distinguished across 8 genera and 12 species, of which 3 were species not native to North America. The most observed bees were *Apis mellifera* (88.3%), *Bombus fervidus* (4.2%), and *B. huntii* (3.7%). To investigate seed production and viability, lavender plants were either caged off and their access withheld from pollinators, or selectively granted access to various pollinators, to determine (1) if lavender is capable of self-pollination, (2) if pollination increases both the seed production rates and seed viability of lavender, and (3) which bee species are the most efficient pollinators of lavender. The findings from this study demonstrate the ecological interactions between pollinators both native and non-native to North America on lavender, a plant native to the Mediterranean region.

**Keywords:** Anthophila; bee; *Lavandula angustifolia*; lavender; pollination; seed



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## 1. Introduction

*Lavandula angustifolia* Mill., lavender, is an aromatic plant in the Lamiaceae (mint) family [1]. While this plant is native to the Mediterranean region, it is cultivated throughout the world, with France and Bulgaria being two of the largest producers [2–4]. Lavender is an important economic plant and crop, prized for its essential oil, and is used extensively in cosmetics, flavors, fragrances, and medicines [5].

Previous studies have shown that lavender is incapable of self-pollination and that, while many insects interact with lavender flowers, bee species are the primary pollinators [6,7]. Multiple studies have been conducted on the relationship between various *Lavandula* species and pollinators [6–12]; however, only a few studies have been conducted on lavender (*L. angustifolia* Mill.). Despite the plant material used (*L. angustifolia*, *L. angustifolia* subsp. *angustifolia*, *L. intermedia*), and the variety of geographic locations (Bulgaria, Algeria, UK) from which previous studies were conducted, findings were similar in that the prominent/proficient pollinators were *Bombus* spp. (followed by *Apis mellifera*) [6–10].

One study found that certain compounds produced by lavender (linalyl acetate, linalyl acetate) were linked to attracting pollinating bees to the flower [6]. Additionally, other studies found that different plant structures of lavender produce different volatile compounds [13–16], and that those found in the calyx are associated with protection from herbivory and those found in the corolla are associated with attraction for pollinators [14].

To the authors' knowledge, only one study has investigated the bee communities that visit lavender in North America [17]. While many different bee species were found visiting

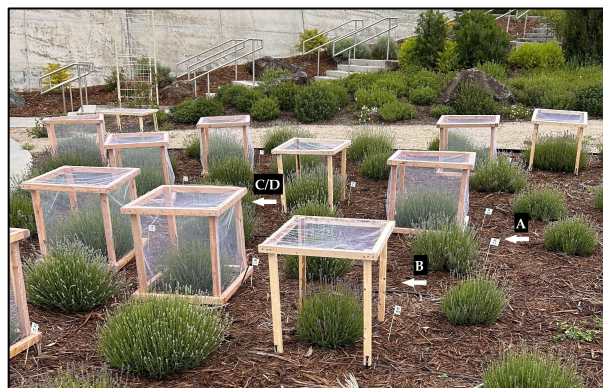
lavender, this study only reported floral visitors and did not investigate how these pollinators affected pollination or seed set [17]. The current study investigates this relationship at a cultivated plot of population lavender (grown from seed) in the state of Utah (USA) during a 4-week flowering period to determine the pollination relationship between pollinators and lavender. To investigate pollination and seed production and seed viability, lavender plants were either withheld from pollinators or pollinators were selectively granted access to determine (1) if lavender is capable of self-pollination, (2) if pollination increases seed production rates and/or seed viability of lavender, and (3) which bee species are the most efficient pollinators of lavender. The findings from this study demonstrate the ecological interactions between lavender, a plant native to the Mediterranean region, and pollinators both native and non-native to North America.

## 2. Material and Methods

The study took place on private land in Lehi, UT, USA (40°25'16" N 111°52'24" W; elevation 1400 m). Two outdoor plots, north and south of a greenhouse edifice, were used for the study. The lavender plants, *Lavandula angustifolia* Mill., were supplied by the Mt. Nebo Botanical Farm (Mona, UT, USA). The Mt. Nebo Botanical Farm cultivates population lavender, which is lavender grown from seed and not from plant cuttings. On each plot, 20 lavender (*Lavandula angustifolia*) plants were chosen at random to be used in the study ( $n = 40$  plants in study). The plants were divided into 4 groups, with half the plants of each group in the north and half in the south plots to mitigate variations in environmental conditions. Group A ( $n = 10$  plants) were uncovered and open to pollination all season (control). Group B ( $n = 10$  plants) were covered on the top of the frames and the sides left open for pollination all season (control). Group C ( $n = 10$  plants) were covered completely and closed off to pollination all season (experimental). Additionally, 50 fine-mesh bags were placed directly over flowering tops and tied at the stem. This system, in the case that self-pollination occurred, would allow for the retention of seeds that are produced and not held within the dried calyx. Group D ( $n = 10$  plants) were covered completely and closed off to pollination, except for select intervals of time, and bees were allowed to visit lavender flowers while under observation (experimental). Once visited, fine-mesh bags were used to cover the flowering tops of the specific flowers that were visited. Fine-mesh bags were collected, for Group C and Group D, on 19 October 2023. When referring to the 4 lavender plant groups, hereafter, they will be referred to as Group A, Group B, Group C, and/or Group D.

Wood frames were built using 5 cm × 5 cm pine material (dimensions h/d/w: 68.5 cm × 61 cm × 61 cm). The covered portions of the wood cages (Group B, Group C, Group D) were covered with fine mesh fabric to exclude pollinators (openings 3 mm × 3 mm), but to allow sun, wind, and rain to penetrate and reach the plants. Mesh was also used on the top of the wood frames as a partial cover for Group B, which was used as a control group (potentially reducing sunlight and wind but still allowing for pollination). Eye screws were put on each 5 cm × 5 cm post at the bottom of the frame to insert stakes to secure the cages to the ground around the plants (Figure 1).

Groups A, B, and D were observed two times a week from 16 June 2023 through to 12 July 2023. Each plant was observed for 15 min increments and picture(s) were taken as insect visitation occurred using a Canon EOS Rebel T6i (Canon Inc., Tokyo, Japan) camera with an attached Canon Macro 100 mm lens. When possible, multiple pictures were taken of each specimen so the insects could be viewed from multiple angles, which aids in identification. Bees and other insect visitors were identified to the lowest taxonomic level from photographs. Identifications were confirmed using online and text resources [18,19]. Group D observations were made as follows: individual contraptions were removed and plants were exposed to pollinators, photos were taken of any visiting bees (as described above), 10 cm × 15 cm fine mesh bags were placed over visited inflorescence, and the original coverings were replaced over the plant.



**Figure 1.** Photo of study plot showing lack of contraction (Group A) and contractions (Group B, Group C, Group D) used for *Lavandula angustifolia* pollination study. Photo taken prior to emergence of corolla (5 June 2023).

Population lavender, which is grown from seed, was used in this study. A representative voucher sample of lavender is held in the Young Living Aromatic Herbarium (YLAH): *Lavandula angustifolia* Mill., Wilson 2023-01 (YLAH).

Seeds from both experimental groups ( $n = 1008$ ) of lavender underwent a 1-month cold stratification process. Seeds were then grown in high-porosity grow media (PRO-MIX HP Mycorrhizae), planted in a greenhouse setting (10 h lighting/day, avg. 25 °C, avg. RH 75%), and allowed to germinate/grow for a 3-month period.

Seedlings are difficult to reliably identify based on morphology alone. To verify that seedlings grown from the collected seeds were lavender, rather than some contaminants in the grow media, dynamic headspace gas chromatography/mass spectrometry (GC/MS) of seedlings' samples ( $n = 10$ ) was conducted using the following instrumentation: a Gerstel Tenax TA desorption liner (Gerstel GmbH & Co. KG, Mülheim, Germany), a Gerstel MultiPurpose Sampler configured with Dynamic Headspace (DHS), a Gerstel Thermal Desorption Unit (TDU 2), a Gerstel Cooled Injection System (CIS 4), Agilent 7890B GC/5977B MSD (Agilent Technologies, Santa Clara, CA, USA), and a Restek Stabilwax (60 m  $\times$  0.25 mm 0.25  $\mu$ m film thickness) fused silica capillary column. Samples were prepared by adding approximately 1 g of lavender plant material (stem, leaf) each to 20 mL screw cap vials (Gerstel), which were capped and placed into the sampling tray. Operating conditions were as follows:

- DHS conditions: vials/samples incubated at 35 °C (1 min), 60 s agitator on time, 1 s agitator off time, 500 rpm agitator speed; 25 °C trapping temp., 3000.0 mL trapping volume, 100.0 mL/min trapping flow.
- TDU 2 conditions: splitless, initial temp. 40 °C; ramp of 720 °C/min, final temp. 280 °C (3 min hold).
- CIS 4 conditions: solvent vent (50 mL/min), split 100:1, initial temp. –40 °C, ramp of 720.0 °C/min, final temp. 280 °C (3 min hold).
- GC/MS conditions: Initial oven temp. at 40 °C (1 min), ramp rate of 3 °C/min, final temp. 240 °C (1 min hold). The electron ionization energy was 70 eV, scan range 40–350 amu, scan rate 4.5 scans per second, source temperature 230 °C, and quadrupole temperature 150 °C. Volatile compounds were identified using the Adams volatile oil library [20] using a Chemstation library search in conjunction with retention indices.

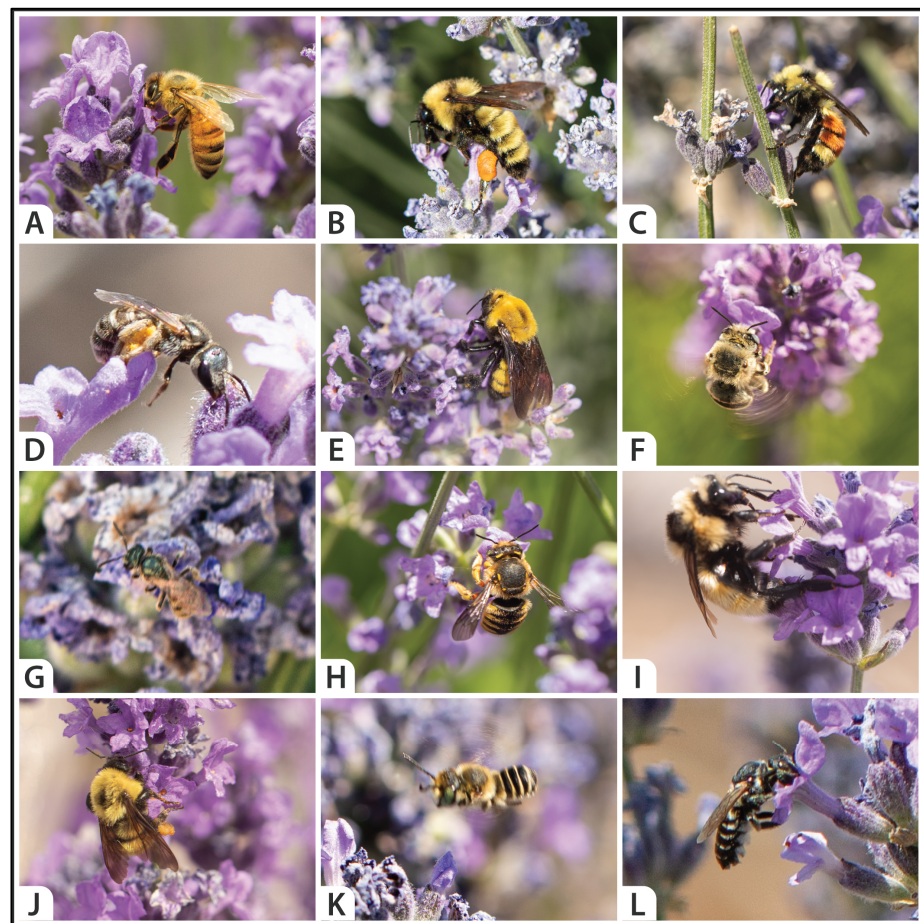
### 3. Results

#### 3.1. Visitation by Bees (Abundance and Distribution)

Over the course of the 4-week observational period, a total of 402 individual bees, distributed among 8 genera and 12 species, were observed foraging on *Lavandula angustifolia* (lavender). While most bees were identified to the species level, two were only identified to



the genus (or subgenus) level. The bees identified include *Apis mellifera*, *Bombus fervidus*, *B. huntii*, *Halictus tripartitus*, *B. morrisoni*, *Anthophora urbana*, *Lasioglossum (Dialictus) sp.*, *Anthidium manicatum*, *B. appositus*, *B. nevadensis*, *Megachile rotundata*, *Hoplitis sp.* (Figure 2). *Bombus* was the most represented genus, which comprised 5 of the 12 bee species observed and identified. Of the bees observed in this study, *A. mellifera*, *B. fervidus*, and *B. huntii* comprised most of the observed bees visiting lavender, over 96%. The exact breakdown is as follows (exact count, rounded %): *Apis mellifera* (355, 88.3%), *Bombus fervidus* (17, 4.2%), *B. huntii* (15, 3.7%), *Halictus tripartitus* (3, 0.7%), *B. morrisoni* (3, 0.7%), *Anthophora urbana* (2, 0.5%), *Lasioglossum (Dialictus) sp.* (2, 0.5%), *Anthidium manicatum* (1, 0.2%), *B. appositus* (1, 0.2%), *B. nevadensis* (1, 0.2%), *Megachile rotundata* (1, 0.2%), *Hoplitis sp.* (1, 0.2%). The complete dataset for bee visitation can be found in Supplementary Table S1.



**Figure 2.** Photos of bees pollinating *Lavandula angustifolia* Mill. (lavender). Identification of bees: (A) *Apis mellifera*, (B) *Bombus fervidus*, (C) *Bombus huntii*, (D) *Halictus tripartitus*, (E) *Bombus morrisoni*, (F) *Anthophora urbana*, (G) *Lasioglossum (Dialictus) sp.*, (H) *Anthidium manicatum*, (I) *Bombus appositus*, (J) *Bombus nevadensis*, (K) *Megachile rotundata*, (L) *Hoplitis sp.* Photos were scaled and cropped in order to best display defining characteristics of bees, however, their actual size can best be determined in reference to the calyx and corolla.

### 3.2. Visitation by Arthropods (Abundance and Distribution)

In addition to the bees observed and identified in the current study, 25 unique Arthropoda were observed. Given that Arthropoda, outside of bees, were not the primary focus of this study, they were identified to the level of order and, when feasible, to the family, genus, and species levels. Some of these Arthropoda are known to be pollinators and others are not, but their interaction with lavender is still noted here. The 25 unique Arthropoda are



represented across nine different orders (Table 1) and the complete dataset for arthropod visitation can be found in Supplementary Table S2.

**Table 1.** Arthropoda (unique Arthropoda and not individuals;  $n = 25$ ) observed interacting with *Lavandula angustifolia*. Those known to be pollinators ( $n = 7$ ) are identified as such. Arthropoda are identified to the order and, when possible, to the species level.

Order	Family	Genus	Species	Pollinator
Araneae	Salticidae	?	?	no
Coleoptera	?	?	?	no
Coleoptera	Melyridae	<i>Collops</i>	?	no
Coleoptera	Lampyridae	?	?	no
Diptera	Muscidae	<i>Coenosia</i>	?	no
Diptera	Syrphidae	?	?	no
Diptera	Syrphidae	<i>Eristalis</i>	<i>tenax</i>	yes
Diptera	Syrphidae	<i>Eupeodes</i>	?	yes
Diptera	Syrphidae	<i>Syritta</i>	<i>pipiens</i>	yes
Diptera	Syrphidae	<i>Toxomerus</i>	<i>marginatus</i>	yes
Diptera	Tachinidae	?	?	no
Diptera	Tephritidae	<i>Trupanea</i>	?	no
Hemiptera	?	?	?	no
Hemiptera	Lygaeidae	<i>Llygaeus</i>	<i>kalmii</i>	no
Hemiptera	Rhopalidae	<i>Liorhyssus</i>	?	no
Hemiptera	Rhopalidae	<i>Liorhyssus</i>	<i>hyalinus</i>	no
Hymenoptera	Braconidae	?	?	no
Hymenoptera	Formicidae	?	?	no
Lepidoptera	Nymphalidae	<i>Vanessa</i>	<i>atalanta</i>	yes
Lepidoptera	Nymphalidae	<i>Vanessa</i>	<i>cardui</i>	yes
Lepidoptera	Pieridae	<i>Pieris</i>	<i>rapae</i>	yes
Neuroptera	Chrysophidae	<i>Chrysoperla</i>	?	no
Odonata	?	?	?	no
Odonata	Coenagrionidae	<i>Enallagma</i>	?	no
Orthoptera	?	?	?	no

### 3.3. Seed Production and Seed Viability

At the end of the growing season, 461 seeds were recovered from Group C lavender plants. There were an estimated 25,000 potential seeds that could be produced from Group C (50 bags, avg. 10 stems/bag, approx. 50 calyx/stem), giving a 1.8% seed production rate. At the end of the growing season, 547 seeds were collected from Group D lavender plants. There were an estimated 17,500 potential seeds that could be produced from Group D (175 bags, avg. 2 stems/bag, approx. 50 calyx/stem), giving a 3.1% seed production rate. The complete dataset for seed viability can be found in Supplementary Table S3.

Group C lavender had a 79.4% seed germination rate and Group D had an 88.3% seed germination rate. To ensure that seedlings were lavender, and not seeds of other species from the growing media, all seedlings ( $n = 849$ ) were visually identified and dynamic headspace GC/MS analysis was performed on select lavender seedlings ( $n = 10$ ) chosen at random, which resulted in the identification of 12 previously determined markers for authentic lavender [16,21].

Group D lavender was used not only to observe which bee species visited lavender, but which bee species were the most successful pollinators of lavender in this study. From that group, 55 of the 175 collected fine-mesh bags contained no seeds, indicating that some visits (about 31% overall) did not result in pollination. *Apis mellifera* was the most common bee species observed (88.3% of the observations). Of the 547 seeds recovered from Group D lavender, 525 were associated with *A. mellifera* and of those, 480 (91.4%) germinated. However, of the 162 observations of *A. mellifera* visiting lavender, only 111 of them resulted in any seed production, meaning only about 68.5% of the visits by *A. mellifera* result in successful pollination. Similarly, only 63.6% (7 of the 11) of the total visits by any *Bombus*

species resulted in successful pollination, but 93.3% of these seeds successfully germinated. When considering all seeds associated with visitations by bee species not native to the Intermountain Region, 68.7% of the visits resulted in pollination and seed set, and 91.2% of those seeds germinated. Similarly, when considering all seeds associated with visitations by bee species that are native to the Intermountain Region, 66.6% of the visits resulted in seed set, and 90.3% of these seeds germinated. Likewise, if you just look at the visits by wild bees (bees other than *A. mellifera*), 69.2% of the visits resulted in seeds, and 86.8% of these germinated.

#### 4. Discussion

*Lavandula angustifolia* (lavender) Group A and Group B were included in the current study to ensure that the presence and diversity of bees was not impacted by the presence of the structures used. Given that Groups A and B were open to pollination throughout the duration of the study (see Material and Methods), these groups were deemed adequate controls and comparisons to each other. The abundance and distribution of the most abundant bees in Groups A and B are comparable (*Apis mellifera* 86.3%, 84.1%; *Bombus fervidus* 6.0%, 8.4%; *Bombus huntii* 2.6%, 2.8%, respectively), with the distribution of other species (<4% of all bees present in the 4-week study) deemed negligible. This supports that the physical contraptions were not a deterrent to visiting pollinators.

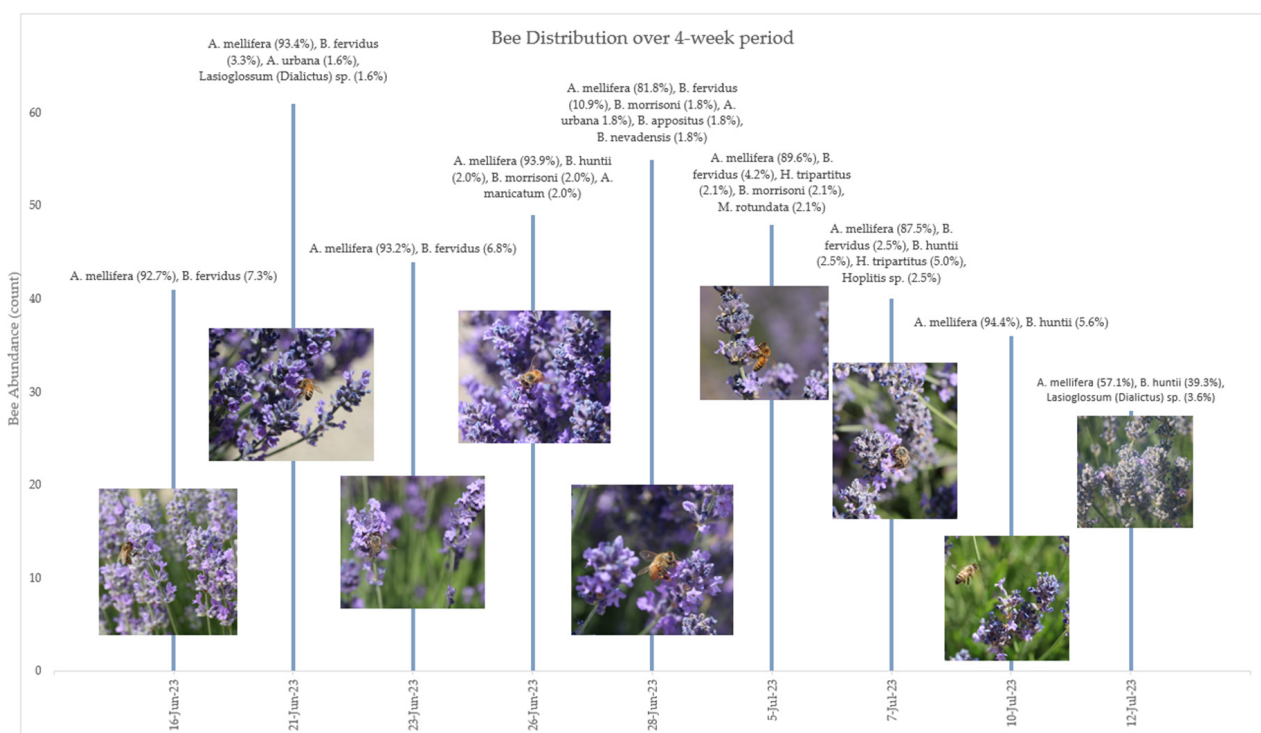
##### 4.1. Visitation by Bees

Of the 12 bee species that were found visiting lavender in the current study, only three species are not native to North America (*Apis mellifera*, *Anthidium manicatum*, *Megachile rotundata*). The three non-native bee species have a large native range, which overlaps parts of the Mediterranean region where lavender originated. In the current study, *A. mellifera* was the most abundant visitor of lavender, representing 88.3% of bee visitation. However, the other two non-native species only represented 0.2% of the bee presence, respectively. Given that the other nine bee species (North American native species) have not historically been exposed to lavender, they could be considered polylectic species (species that collect pollen from a variety of plants). Most of the bee species we observed visiting lavender are known polylectic species, with one possible exception, the *Hoplitis* species. Many species of *Hoplitis* exhibit a preference for specific plant families or even plant genera. As it was not possible to identify the species from the photos, it is unknown if this species was a specialist or a generalist bee.

Similar studies to this topic, performed in Bulgaria and Algeria, also found that bees were the prominent pollinators of lavender (compared to butterflies, moths, etc.) [7,10]. Contrary to the current study, previous research found that *Bombus* spp. were both the most prominent and most effective pollinators, and that they were able to outcompete *A. mellifera* [7–10]. In the Intermountain Region (USA), *Bombus* spp. are most abundant and diverse in mountains [22,23], while the current study was performed in a valley floor of the Great Basin (an area where pollinators are greatly influenced by non-native plants, agriculture, horticulture, etc.). Balfour and associates, who researched the relationship between bees and *Lavandula intermedia*, credited the prominence of *Bombus* spp. (approx. 1:9 *Apis* sp. to *Bombus* spp.) to the longer tongue length (7.8–8.9 mm) compared to *A. mellifera* (approx. 6.6 mm) [8]. Since the length of the corolla tube is approximately 7 mm [8], *Bombus* spp. can access nectar from various angles while *A. mellifera* are forced to insert their face into the corolla. While the cause for the inverse ratio (approx. 10:1) of *A. mellifera* to *Bombus* spp. in the current study may be due to location of the study (both lavender and *A. mellifera* co-evolved in Mediterranean region), this is mainly speculation and additional research is needed to investigate this cause. Our results, however, are similar to another recent study of bee communities visiting lavender in North America, that found *A. mellifera* was the most abundant visitor of the plant [17].

Throughout the 4-week observational period spanning nine different days, the fluctuation in the overall abundance and diversity of bees pollinating lavender changed (Figure 3).

*Apis mellifera* was the only bee species that was observed visiting lavender on every day of observation, comprising between 57.1–94.4% of the bees present on any given single day. The second most abundant bee species, *B. fervidus*, was observed throughout most of the study (6 of 9 days). The third most abundant bee species, *B. huntii*, was observed almost exclusively at the end of the study, particularly the final day of observation (11 of the 15 observations) as corolla senescence was prominent. There appears to be a parallel relationship between the total number of pollinators observed each day and the level of lavender maturity depicted by the abundance of open corolla. Lavender, during the beginning of the observational period, was less mature (majority of corolla yet to emerge), had a peak of maturation in the middle of the study (26 June, 28 June), and began to senesce by the end of the study (10 July, 12 July) (Figure 3). However, this period of senescence also coincides with the prominent presence of *B. huntii*, suggesting that this species was either previously outcompeted by or is less selective than *A. mellifera*. Li and associates suggest the prolonged flowering and pollination period of lavender, described as the “sequential and successive [bloom]”, is responsible for the extended presence of pollinators like *B. huntii* [6]. Additionally, researchers in Portugal determined that bees were selective foragers of *Lavandula stoechas*, based on the number of open corolla and nectar content [11]. While both findings are confirmed by the current study, it is important to emphasize that the visitation of lavender by bee species is not a straightforward or linear trend. Instead, it varies from species to species and does not appear to conform to a general pattern of foraging behavior and timing among all bee species. Each bee species may have its unique preferences and behaviors when interacting with lavender.



**Figure 3.** Timeline of the abundance of total bees observed each day of observation (*y*-axis) over a 9-day (extending over 4 weeks) period (*x*-axis). Each day of observation is further distinguished by the flowering stage of *Lavandula angustifolia* (lavender) and the distribution (%) of bees species observed.

#### 4.2. Visitation by Arthropods

Arthropoda, apart from bees, were not the focus on the current study. However, it is important to understand how Arthropoda, both native and non-native to the Intermountain Region, interact with a non-native plant species that is broadly cultivated throughout North America. In the current study, including bees, 37 Arthropoda species, largely native to this



region, were observed interacting (foraging, herbivory, shelter, and/or insectivory) with a non-native plant species, lavender. Future studies should expend additional time and resources on this interaction, as it would provide valuable insights into the broader impact of lavender cultivation and the various roles Arthropoda play in these interactions.

#### 4.3. Seed Production and Seed Viability

Given the seed production ( $n = 461$ ) and seed viability (79.4% germination rate) from Group C lavender plants, self-pollination of lavender appears to occur. Previous research found that lavender is incapable of self-pollination [7]. However, while both studies were conducted on lavender (*Lavandula angustifolia*), the previous study [7] was conducted on lavender produced by clonal propagation and the current study was conducted on population lavender, grown from seed. Since the larger contraption netting used in the current study contained openings  $3\text{ mm} \times 3\text{ mm}$  and the fine-mesh bags contained openings  $1\text{ mm} \times 1\text{ mm}$ , it is extremely unlikely that pollinating insects were responsible for seed production. Future studies could further investigate if different cultivars of lavender, or if lavender produced by different cultivation methods (clonal cuttings vs. population), are capable of self-pollination.

Additionally, the 72% increase in seed viability from Group C to Group D lavender (1.8% to 3.1%) suggests that even a single pollination event of lavender by bees leads to higher seed viability and germination rates. Overall, there was not a big difference in the pollination rate or rate of seed germination between *A. mellifera* and wild bees, though some of the wild bees were more effective pollinators than others. Of the 12 bee species observed visiting lavender, we found that four species can be classified as good pollinators of lavender, some native to the overlapping native habitat of lavender (*Apis mellifera*, *Anthidium manicatum*) and others native to North America (*Bombus fervidus*, *B. huntii*). It should be noted that since self-pollination does occur, it is difficult to determine what actual percent of seeds from Group D lavender were produced from self-pollination vs. insect–plant interactions. Additionally, from Group D lavender, 31% (55 of 175) of the fine-mesh bags contained no seeds, suggesting that some bee species are “nectar robbing” or are otherwise inefficient pollinators of lavender.

Seed viability research is commonly conducted on sterile grow media, such as paper or agar bases [24,25]. Using non-sterile grow media in seed viability studies may cause issues if the media are contaminated with seeds. Using traditional taxonomic methods for identifying germinating seedlings proves difficult when identifying plants to the species level (lack of reproductive parts, etc.) or, at times, distinguishing the seedlings of interest from possible contaminants in the grow media. In the current study, all seedlings ( $n = 849$ ) were identified visually (dicotyledon, leaves opposite, etc.). Additionally, the seedlings of randomly selected plants ( $n = 10$ ) were analyzed by dynamic headspace GC/MS to identify volatile compounds produced by the lavender seedlings that are key markers for that plant species. While some seedling dynamic headspace GC/MS samples contained additional markers, all ten samples contained the same 12 compounds ( $\alpha$ -thujene,  $\alpha$ -pinene,  $\delta$ -3-carene, limonene, camphor, borneol, geranyl acetate,  $\alpha$ -santalene, (E)-caryophyllene,  $\gamma$ -cadinene, caryophyllene oxide, epi- $\alpha$ -cadinol) that have previously been identified as markers for authentic lavender [16,21]. Some key volatile compounds, typically associated with mature lavender (linalool, linalyl acetate), were not detected in all seedlings analyzed by dynamic headspace GC/MS. However, this appears to be consistent with the previous findings that plants at different growth stages produce different volatile compounds [13–16,26]. The complete quantitative dataset for dynamic headspace GC/MS can be found in Supplementary Table S4.

## 5. Conclusions

Lavender (*Lavandula angustifolia*) is a perennial flowering plant that is cultivated throughout the world. The current study is the first of its kind to investigate which bee

species visit and pollinate cultivated lavender in North America, and to investigate the impact of pollination on seed production and seed viability.

The current study concluded that 12 bee species were found to visit cultivated lavender in the Intermountain Region of North America and that while some bee species may be “nectar robbing”, or otherwise inefficient pollinators of lavender, four species can be identified as good pollinators of lavender (*Apis mellifera*, *Anthidium manicatum*, *Bombus fervidus*, *B. huntii*). The current study also found that while self-pollination of population lavender does occur, pollination by various bee species increases both seed production rates and seed viability. When identifying seedlings, dynamic headspace GC/MS analysis was conducted on the volatile compounds produced by lavender. This method proves to be a novel and reliable technique to identify aromatic plant species when reproductive plant parts are lacking due to plant age and maturity.

This study brings to light the ecological findings between both native and non-native bees, and other Arthropoda, on a non-native plant species, lavender, in North America. While the cultivation of plants at either small or large scale could be viewed as a trivial practice, its impact on the local flora and fauna can be substantial. Given constant environmental changes, both natural and influenced by humans, future studies should investigate how these changes impact insect pollinator interactions. For instance, what are the broader impacts of introducing non-native plant species to new regions, how are insect pollinators adapting to ecological stresses, and/or how are plants adapting to environmental changes? These findings could bring value to an ever-changing world and could bring insights into improving agricultural techniques as we face new challenges with ensuring food security. Additionally, future studies should investigate if findings from a mirrored study are the same from year-to-year or differ in other geographical locations and environments.

**Supplementary Materials:** The following supporting information can be downloaded at: <https://www.mdpi.com/article/10.3390/seeds3020021/s1>, Table S1: Bee Data; Table S2: Arthropod Data; Table S3: Seed Data; Table S4: Dynamic Headspace GC/MS Data.

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**Data Availability Statement:** The data presented in this study are available upon request from the corresponding author.

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## References

1. The World Flora Online. Available online: <https://www.worldfloraonline.org/taxon/wfo-0000224116> (accessed on 9 September 2023).
2. Bailey, L.H. *Manual of Cultivated Plants*, revised ed.; Macmillan Publishing Company: New York, NY, USA, 1951; pp. 850–851.
3. Lis-Balchin, M. *Lavender: The Genus Lavandula*, 1st ed.; Taylor & Francis: London, UK, 2002; pp. 35–45.
4. Stanev, S.; Zagorcheva, T.; Atanassov, I. Lavender cultivation in Bulgaria—21st century developments, breeding challenges and opportunities. *Bulg. J. Agric. Sci.* **2016**, *22*, 584–590.

5. Khan, S.U.; Hamza, B.; Mir, R.H.; Fatima, K.; Malik, F. Lavender plant: Farming and Health benefits. *Curr. Mol. Med.* **2023**, *24*, 702–711. [[CrossRef](#)] [[PubMed](#)]
6. Li, H.; Li, J.; Dong, Y.; Hao, H.; Ling, Z.; Bai, H.; Wang, H.; Cui, H.; Shi, L. Time-series transcriptome provides insights into the gene regulation network involved in the volatile terpenoid metabolism during the flower development of lavender. *BMC Plant Biol.* **2019**, *19*, 313. [[CrossRef](#)]
7. Valchev, H.; Kolev, Z.; Stoykova, B.; Kozuharova, E. Pollinators of *Lavandula angustifolia* Mill., an important factor for optimal production of lavender essential oil. *BioRisk* **2022**, *17*, 297–307. [[CrossRef](#)]
8. Balfour, N.J.; Garbuzov, M.; Ratnieks, F.L. Longer tongues and swifter handling: Why do more bumble bees (*Bombus* spp.) than honey bees (*Apis mellifera*) forage on lavender (*Lavandula* spp.)? *Ecol. Entomol.* **2013**, *38*, 323–329. [[CrossRef](#)]
9. Balfour, N.J.; Gandy, S.; Ratnieks, F.L. Exploitative competition alters bee foraging and flower choice. *Behav. Ecol. Sociobiol.* **2015**, *69*, 1731–1738. [[CrossRef](#)]
10. Benachour, K. Insect visitors of lavender (*Lavandula officinalis* L.): Comparison of quantitative and qualitative interactions of the plant with its main pollinators. *Afr. Entomol.* **2017**, *25*, 435–444. [[CrossRef](#)]
11. Duffield, G.E.; Gibson, R.C.; Gilhooly, P.M.; Hesse, A.J.; Inkley, C.R.; Gilbert, F.S.; Barnard, C.J. Choice of flowers by foraging honey bees (*Apis mellifera*): Possible morphological cues. *Ecol. Entomol.* **1993**, *18*, 191–197. [[CrossRef](#)]
12. Herrera, C.M. Daily patterns of pollinator activity, differential pollinating effectiveness, and floral resource availability, in a summer-flowering Mediterranean shrub. *Oikos* **1990**, *58*, 277–288. [[CrossRef](#)]
13. Adaszynska-Skwirzynska, M.; Dzieciol, M. Comparison of chemical composition and antimicrobial activity of essential oils obtained from different cultivars and morphological parts of *Lavandula angustifolia*. *J. Essent. Oil-Bear. Plants* **2018**, *21*, 1532–1541. [[CrossRef](#)]
14. Guitton, Y.; Nicolè, F.; Moja, S.; Valot, N.; Legrand, S.; Jullien, F.; Legendre, L. Differential accumulation of volatile terpene and terpene synthase mRNAs during lavender (*Lavandula angustifolia* and *L. x intermedia*) inflorescence development. *Physiol. Plant.* **2010**, *138*, 150–163. [[CrossRef](#)] [[PubMed](#)]
15. Özel, A. Determining leaf yield, some plant characters and leaf essential oil components of different cultivars of lavender and lavandin (*Lavandula* spp.) on the Harran plain ecological conditions. *Appl. Ecol. Environ. Res.* **2019**, *17*, 14087–14094. [[CrossRef](#)]
16. Wilson, T.M.; Poulson, A.; Packer, C.; Carlson, R.E.; Buch, R.M. Essential oil profile and yield of corolla, calyx, leaf, and whole flowering top of cultivated *Lavandula angustifolia* Mill. (Lamiaceae) from Utah. *Molecules* **2021**, *26*, 2343. [[CrossRef](#)] [[PubMed](#)]
17. Wilson, J.S.; Young, J.G.; Wilson, L.T. The Bee Communities of Young Living Lavender Farm, Mona, Utah, USA. *Diversity* **2024**, *16*, 119. [[CrossRef](#)]
18. iNaturalist. Available online: <https://www.inaturalist.org> (accessed on 17 July 2023).
19. Carril, O.M.; Wilson, J.S. *Common Bees of Western North America*; Princeton University Press: Princeton, NJ, USA, 2023.
20. Adams, R.P. *Identification of Essential Oil Components by Gas Chromatography/Mass Spectrometry*, 4th ed.; Allured Publ.: Carol Stream, IL, USA, 1997.
21. Wilson, T.M.; Poulson, A.; Murphy, B.J.; Nebeker, B.; Cuchet, A.; Schiets, F.; Casabianca, H.; Carlson, R.E. Authentication of *Lavandula angustifolia* Mill. (Lamiaceae) essential oil using physical property, gas chromatography, enantiomeric selectivity, and stable isotope analyses. *J. Essent. Oil Res.* **2023**, *35*, 529–541. [[CrossRef](#)]
22. Williams, P.H.; Thorp, R.W.; Richardson, L.L.; Colla, S.R. *Bumble Bees of North America: An Identification Guide*; Princeton University Press: Princeton, NJ, USA, 2014; Volume 87.
23. Wilson, J.S.; Pan, A.D.; Alvarez, S.I.; Carril, O.M. Assessing Müllerian mimicry in North American bumble bees using human perception. *Sci. Rep.* **2022**, *12*, 17604. [[CrossRef](#)] [[PubMed](#)]
24. Esfandiari, A.; Norling, C.; Kaji, R.; McLachlan, A.; Mathew, L.; Fleming, M.; Morgan, E.; Nadarajan, J. Variations in Seed Dormancy Occurrence and Their Classifications in Thirteen *Actinidia* Species. *Seeds* **2024**, *3*, 179–195. [[CrossRef](#)]
25. Silva, J.D.J.; Gomes, R.A.; Ferreira, M.A.R.; Pelacani, C.R.; Dantas, B.F. Physiological Potential of Seeds of *Handroanthus spongiosus* (Rizzini) S. Grose (Bignoniaceae) Determined by the Tetrazolium Test. *Seeds* **2023**, *2*, 208–219. [[CrossRef](#)]
26. Détár, E.; Németh, É.Z.; Gosztola, B.; Demján, I.; Pluhár, Z. Effects of variety and growth year on the essential oil properties of lavender (*Lavandula angustifolia* Mill.) and lavandin (*Lavandula x intermedia* Emeric ex Loisel.). *Biochem. Syst. Ecol.* **2020**, *90*, 104020. [[CrossRef](#)]

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