Life cycle and reproductive botany of Scilla hyacinthoides, a Mediterranean geophyte

ARTICLE in SCIENTIA HORTICULTURAE · MARCH 2016
Impact Factor: 1.37 · DOI: 10.1016/j.scienta.2016.01.043

3 AUTHORS:

Ilana Shtein
Hebrew University of Jerusalem
9 PUBLICATIONS  13 CITATIONS
SEE PROFILE

Tal Noy-Porat
Tel Aviv University
10 PUBLICATIONS  40 CITATIONS
SEE PROFILE

Amram Eshel
Tel Aviv University
76 PUBLICATIONS  1,956 CITATIONS
SEE PROFILE

All in-text references underlined in blue are linked to publications on ResearchGate, letting you access and read them immediately.

Available from: Ilana Shtein
Retrieved on: 19 March 2016
Life cycle and reproductive botany of *Scilla hyacinthoides*, a Mediterranean geophyte

Ilana Shtein a,*,1, Tal Noy-Porat b,1, Amram Eshel c

a The Robert H. Smith Institute of Plant Sciences and Genetics in Agriculture, The Hebrew University of Jerusalem, The Robert H. Smith Faculty of Agriculture, Food & Environment, Rehovot 76100, Israel

b The Department of Biochemistry, Israel Institute for Biological Research, Nes Ziona, Israel

c The Department of Molecular Biology and Ecology of Plants, Tel Aviv University, Tel Aviv, Israel

**A R T I C L E   I N F O**

Article history:
Received 21 April 2015
Received in revised form 20 January 2016
Accepted 25 January 2016

Keywords:
Florogenesis
Flower polymorphism
Temperature
Anatomy

**A B S T R A C T**

The Mediterranean region is abundant with geophyte species, many of which have been developed as ornamentals. In recent years attempts were made to grow *Scilla hyacinthoides* L. (Asparagaceae, subfamily Scilloideae) commercially, both as a cut flower and as a water-saving ornamental geophyte. Optimizing commercial production of *S. hyacinthoides* requires the study of growth and development of the vegetative plant, as well as the control of flower bud initiation and development.

Like most Mediterranean plants, *S. hyacinthoides* life cycle is attuned to the annual rhythm which alternates between hot and dry summer, and cool and rainy winter. Plants are dormant during summer. In autumn the apical meristem becomes active and either begins producing leaves for the next season, or initiates inflorescence development. After the onset of inflorescence development, a new apical meristem is formed at the axil of the youngest leaf. Frequently, two apices are formed; one of them becomes a daughter bulb. Intra-bulb development of the inflorescence was studied by histology and electron microscopy. The flowering occurs in spring. Floral initiation is temperature-dependent and is inhibited by low temperatures, but inflorescence differentiation and elongation is inhibited by high temperatures. Flowers are self-incompatible and exhibit several polymorphic traits, probably important in pollinator attraction. In summary, current research provides a basis for future studies on the florogenesis and ecophysiology in *Scilla*, as well as enabling future fine-tuning of cultivation protocols for the flower industry.

© 2016 Elsevier B.V. All rights reserved.

1. Introduction

*Scilla hyacinthoides* L. (Asparagaceae, subfamily Scilloideae) is a perennial bulbous geophyte, native to the Mediterranean region—from Portugal to Israel, Iraq and south-east Turkey (Halevy, 1985). According to Wirtzum and Negbi (1991), *S. hyacinthoides* is the “wool-bearing bulb” mentioned by the father of botany, Theophrastus: when bulb scales of *S. hyacinthoides* are transversely pulled apart, many woolly threads are seen. The “wool” is comprised of stretched helical secondary walls of the primary xylem cells. Theophrastus described a use for these threads in making “felt shoes and other articles of apparel”.

The Mediterranean climate is characterized by two distinct seasons: a cool, rainy winter and a hot and dry summer. Therefore, the Mediterranean plants are usually physiologically active from autumn to spring and dormant during the summer. Such adaptation determines their structure and annual developmental cycles. This is especially true for geophytes. Geophytes possess subterranean storage organs with renewal buds that are located below the soil surface. At the beginning of the dry summer season the geophytes enter a state of dormancy, during which the plants do not exhibit any visible external growth. However, complex processes occur within the geophyte storage organs during dormancy period, such as organogenesis, development of newly formed flowers and leaves and degradation of the storage materials (Kamenetsky, 2009).

Mediterranean geophytes can be divided into two groups based on their life cycle: (1) synanthous geophytes, in which leaves and flowers appear immediately after one another and the course of events is growth, storage, flowering and dormancy and (2) hysteronanthous geophytes, in which flowers and leaves appear separately and the course of events is growth, storage, dormancy and flowering (Dafni et al., 1981a). *S. hyacinthoides* belongs to the synanthous group. In summer the leaves dry out and the bulb remains dormant.

* Corresponding author.
  
  E-mail addresses: Ilana.Shtein@mail.huji.ac.il, itisilana@hotmail.com (I. Shtein).
  
  1 The first two authors are joint authors of this article.

http://dx.doi.org/10.1016/j.scienta.2016.01.043
0304-4238/© 2016 Elsevier B.V. All rights reserved.
in the ground. The bulb resprouts in autumn (September–October) and in Israel the blooming occurs in spring (March–April). In the Northern Mediterranean, where winter is colder, the leaf growth and flowering are delayed, accordingly, to spring and early summer (Tutin et al., 1980). The course of S. hyacinthoides annual development, and inflorescence differentiation stages have never been described.

Flowering in geophytes is divided into several stages: induction, initiation, differentiation, maturation and growth of the organs, anthesis and senescence (Kamenetsky et al., 2012). Each stage is regulated by numerous internal and external factors. Among the external environmental factors (light, moisture, temperature), temperature is considered the principal factor of geophytes growth and flowering (Khodorova and Boitel-Conti, 2013). Floral induction takes place entirely at the molecular level, and cannot be detected morphologically. The first signs of morphological changes in the meristem mark the initiation step. These changes include the enlargement of the meristem, which becomes dome-shaped. The former vegetative meristem is now an inflorescence meristem, which gives rise to individual flower primordia. Next, the individual flower organs are developed, during the differentiation step. Flowering stages of numerous ornamental geophytes has been described in detail, as such knowledge is essential in geophyte flower forcing (for example, tulip, Gilford and Rees, 1974; Allium rothii, Kamenetsky, 1994; Narcissus tazetta, Noy-Porat et al., 2009).

Most currently grown “classic” bulbous ornamental plants originated from the Middle East and Central Asia, such as anemone, ranunculus, cyclamen, hyacinth, N. tazetta, Lilium candidum and several tulips (Halevy, 2000), and many more attractive species can be found in these regions. Species belonging to the genus Scilla have a great ornamental potential, but only few are grown commercially and only little is known about their botany and life cycle (Azizbekova et al., 1998). S. hyacinthoides has showy inflorescences with tall stalks, bearing over one hundred blue–violet flowers. In the recent years attempts were made to grow S. hyacinthoides commercially both as a cut flower and as a water-saving ornamental geophyte (Shtein et al., 2014). However, little is known about its reproductive botany, which is crucial for further cultivation attempts. A detailed cytological and morphological study is needed for development of an effective commercial forcing program for S. hyacinthoides. The current research attempts to close this gap.

2. Materials and methods

2.1. Growth parameters and morphology

Seventy mature bulbs were planted, during early autumn, at the Tel Aviv University Campus, in soil. Each month, 3–5 bulbs were randomly harvested. Bulb diameter, weight and leaf number were determined and the bulbs were dissected to determine their morphology and the timing of initiation and development of different organs around the year. During the months of flower development bulbs were examined every week. Shoot apices were isolated from these bulbs for anatomical examination of the apical meristem. In addition, inflorescences were screened for flower polymorphism.

2.2. Bulb structure

Mature bulbs were carefully dissected scale after scale to determine numerous parameters, including number of scales, position and number of inflorescences, position and number of daughter bulbs and the state of the apical meristem.

2.3. Root length measurement

Roots were cut off from each bulb, rinsed with tap water, and immersed in neutral red solution (0.2 g/L) over night. The next day, the roots were rinsed again and dispersed randomly in a shallow water bath. Roots were then scanned in an Expression 1600 scanner (Epson) and roots total length and diameter were analyzed by WinRhizo (Regent Instruments Inc.). Number of roots for each bulb, and the formation of lateral roots were also recorded.

2.4. The temperature effect on the apical meristem

The effect of temperature on apex differentiation was tested on mature bulbs planted in 2L pots filled with potting mixture and placed in controlled growth chambers with constant temperature of 15 °C, 25 °C, and 30 °C (12 h photoperiod). The pots were irrigated with tap water three times a week.

2.5. Pollination

Mature bulbs were planted in soil, in autumn, at the Tel Aviv University Campus. Two pollination experiments were performed. In the first experiment a group of inflorescences was covered with transparent mesh bags, and uncovered inflorescences served as control. In the second experiment all the inflorescences were covered with transparent mesh bags. The inflorescences were divided into two treatments: one was cross-pollinated (between flowers of different plants) by hand using a paint brush, and the other was self-pollinated (between flowers of the same inflorescence) the same way. Two months later the number of fertilised flowers which formed immature fruits was counted.

2.6. Histology

Plant material was fixed in FAA (5:5:90, formaline: acetic acid: 70% ethanol), dehydrated in a graded alcohol series and then embedded in paraffin wax. Cross-sections of 10 μm thickness were cut with a rotary microtome and stained with safranin-fast green. The sections were viewed under a light microscope (Nikon Eclipse E600) equipped with a camera (Nikon Coolpics 995).

2.7. SEM

Apices at different stages were isolated and prepared for scanning electron microscopy (SEM) (according to Garty and Delarea, 1987). The samples were viewed and photographed with a Jeol-840A SEM.

2.8. Statistical analysis

Statistical differences were examined using one-way ANOVA analysis of variance, followed by Tukey post-hoc test for statistically different treatments. Analysis was performed with Statistica 6 software (StatSoft, Inc.). The data was not transformed since no deviation from normality was observed.

3. Results

3.1. Mature bulb morphology and structure

The bulb of S. hyacinthoides is composed of a basal plate and scales that are bases of the leaves of the previous two years (Fig. 1C). Average mature bulb circumference is 19.5 cm (Table 1). A mature bulb produces each season 10–14 green leaves (Table 1). The glabrous leaves of light green color are linear—lanceolate, 30 cm long and 3–5 cm wide. When grown in an open space they form a
prostrate rosette. When growing in clumps or within dense vegetation they assume various higher angles. During the growth season the leaf bases accumulate water and storage compounds and become swollen. At the end of the season the leaf laminae dry up and the bases remain in the bulb, turning into the bulb scales. The scales surround the apex and protect it. Older scales empty and form a multilayered tunica, which protects the outer surface of the bulb. As the bulb grows, the outer tunica layers are shed and the inner scales form new tunica layers.

The bases of inflorescence scapes of previous years could be found among the scales. The average number of scales between two subsequent inflorescences was not significantly larger than the annual leaf number (17 versus 12, Table 1). Mature bulbs of *S. hyacinthoides* often produce daughter bulbs between the scales. They take two years until they reach the periphery of the bulb and grow into independent offshoots. Usually only one or two daughter bulbs are formed per mature bulb annually (1.6 ± 2.1 daughter bulbs were found per bulb, Table 1). Daughter bulbs were usually formed in the proximity of an inflorescence (Fig. 1). Sometimes a larger number (5–10) of small offshoots developed on the periphery of the bulb at the same time.

3.2. The annual life cycle

3.2.1. Autumn

The growth season begins with emergence of preformed leaves and the initiation of new roots. The leaves grow rapidly, form the large rosette, and supply photosynthates for the seasonal growth. Roots begin to appear after the first rains. The roots, which are formed around the basal plate, are contractile, branched and have root hairs.

The renewed activity in the apical meristem starts with increased activity in the nuclei of the meristematic cells throughout the meristem (data not shown). The development of the apex may take one of two directions: it either remains vegetative or turns reproductive. Meristematic activity of the vegetative apex leads to formation of new leaves. The apex continues to form leaves throughout the growing season until spring (Fig. 2). These newly formed leaves will appear aboveground the next year. Reproductive development will lead to the formation of an inflorescence and to the development of a new vegetative apex, which appears at the axil of the youngest leaf that was formed the year before (Fig. 3B–D). This new apical meristem starts forming leaves that will also appear aboveground in the next year.

The first visible sign of the reproductive differentiation is an increase in the size of the meristem, which becomes dome-shaped (Fig. 3A,E). The flower primordia develop acropetally (Fig. 3A–D). The apex continues to elongate while more flower primordia appear. Eventually all flowers in the inflorescence are formed and there is a clear distinction between the scape and the part of the inflorescence that bears the flowers. At the same time, the flower organs differentiate as well (Fig. 3F–H). First, the bract primordium appears at the base of the flower primordium (Fig. 3F). Next, the first whorl of three sepals is formed and then a second whorl of petals (Fig. 3G). In the next stages six stamens and gynoecium are formed (Fig. 3H). At each stage, the lowest flowers are the most differentiated. The inflorescence differentiation takes approximately two weeks.

3.2.2. Winter

During winter the inflorescence grows and develops inside the bulb, while the vegetative apex continues to form leaves. The inflorescence elongates and by the end of February emerges from the bulb. By this time, the vegetative apex had initiated 7 or 8 leaf primordia (Fig. 2C).

3.2.3. Spring

Anthesis takes place from early March to early April. In the first ten days there is a rapid elongation of the scape, above ground. Afterwards the flowers open in an acropetal order, in parallel with the stem elongation. The inflorescence is eventually around 1 m tall and is composed of 50–150 flowers. The flowers are visited by bees and many other insects. At the end of flowering, fruits develop and the seeds mature. The vegetative apex remains active (Fig. 2D).

3.2.4. Summer

Leaves and most of the roots dry up gradually and the bulb enters a state of dormancy. Some roots remain alive and turgid. The fruits dry up and are dispersed during the summer through the movements of the erect scape. The vegetative apex becomes inactive (Fig. 2A).

Table 1

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Average</th>
<th>St.Dev (N = 26)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bulb circumference (cm)</td>
<td>19.48</td>
<td>1.62</td>
</tr>
<tr>
<td>Scale N per season</td>
<td>12.88</td>
<td>1.98</td>
</tr>
<tr>
<td>N of scales between two subsequent inflorescences</td>
<td>17.63</td>
<td>5.84</td>
</tr>
<tr>
<td>Daughter bulbs N</td>
<td>1.61</td>
<td>2.09</td>
</tr>
<tr>
<td>Inflorescence scapes N</td>
<td>1.31</td>
<td>0.75</td>
</tr>
</tbody>
</table>

Fig. 1. Occurrence of daughter bulbs within the dissected bulbs. (A) Position of the daughter bulbs (dissected bulb N = 26). (B) A daughter bulb located near a dry inflorescence scape exposed by removal of part of the mature bulb scales. Is—inflorescence scape, Db—daughter bulb. C—Schematic representation of a mature *S. hyacinthoides* bulb.
Fig. 2. Vegetative apical meristem activity during the year. (A) Summer, (B) Autumn, (C) Winter, (D) Spring. AM—apical meristem, LP—leaf primordia.

Fig. 3. Inflorescence and flower development. (A–D) Light microscope images of several stages of the reproductive meristem development into an inflorescence. (E–H) SEM images of several stages of individual flower development. Arrows indicate individual flowers. AM—apical meristem, IP—inflorescence primordium, FP—Flower primordium, B—bract, TP—tepall primordium.
3.3. Effect of growth temperature on inflorescence initiation and development

Growth temperature had a strong impact on initiation of flowering (Fig. 4A). At 25 °C initiation of the inflorescence started about a month earlier than in the other two temperatures and reached high initiation percentages (about 80%). Bulbs at 30 °C showed similar degree of initiation of about 80%, while at 15 °C the percentage of initiation was very low (about 40%).

However, temperature had a different effect on inflorescence differentiation. At 15 °C there was very low percentage of floral initiation, but at the end of the experiment those inflorescences that did develop reached the biggest size (Fig. 4B). At 30 °C, though initiation percentages were high, the differentiation was severely inhibited at an early stage (Fig. 4B,C) and eventually the inflorescences did not develop fully.

Growth temperature also affected leaf emergence and root development. Growth at 30 °C resulted in severe inhibition of leaf emergence (Table 2). Less than 10% of the bulbs showed leaf emergence which was also extremely delayed compared to the other temperatures. Root number decreased with increase of temperature (Table 2). Roots were also shorter at 30 °C, compared to the other temperatures. In addition, the root system was much more branched in bulbs grown at 15 °C or 25 °C, while very few adventive roots developed at 30 °C (data not shown). The average root diameter was lower in 25 °C, compared to the other temperatures (Table 2). This is due to a much more branched root system and formation of many thin side roots in this temperature. As expected, growth temperature had no effect on leaf number, since they were formed in the previous year. Bulb size was also not affected by temperature.

3.4. Self-incompatibility

In order to assess S. hyacinthoides ability to self-fertilize, two self-pollination experiments were performed. In the first experiment the treatment group was covered with transparent mesh bags to prevent pollination by insects; uncovered inflorescences served as control. Both control and treatment inflorescences had about 200 flowers (results not shown). The percentage of fertilized flowers in the covered inflorescences was much lower (5%) than in the control, uncovered ones (57%) (Fig. 5A).

In the second experiment all the inflorescences were covered with transparent mesh bags. The inflorescences were divided into two groups: one cross-pollinated by hand, and the other one self-pollinated by hand. The cross-pollinated flowers had 89% of fertilization as compared to 13% fertilization in self-pollinated ones (Fig. 5B).

Table 2
Temperature effect on plant emergence and development. Each measurement is an average of 30 bulbs examined, differences between temperatures were determined by one-way ANOVA and Tukey post-hoc test (p < 0.05).

<table>
<thead>
<tr>
<th>Growth temperature</th>
<th>Bulb diameter (cm)</th>
<th>Bulb Weight (g)</th>
<th>Leaf N</th>
<th>(%) Leaf emergence</th>
<th>Time of leaf emergence</th>
<th>Root N</th>
<th>Root length (mm)</th>
<th>Root diameter (mm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>15 °C</td>
<td>5.71 ± 0.67</td>
<td>116.7 ± 34.5</td>
<td>11.48 ± 2.25</td>
<td>100</td>
<td>1–15 Oct</td>
<td>27.9 ± 8.9</td>
<td>789.2 ± 279.7</td>
<td>1.39 ± 0.18</td>
</tr>
<tr>
<td>25 °C</td>
<td>5.55 ± 0.79</td>
<td>102.5 ± 34.7</td>
<td>11.22 ± 2.76</td>
<td>100</td>
<td>1–10 Oct</td>
<td>25.4 ± 7.6</td>
<td>670.1 ± 330.4</td>
<td>1.17 ± 0.14</td>
</tr>
<tr>
<td>30 °C</td>
<td>5.91 ± 0.59</td>
<td>115.4 ± 31.6</td>
<td>11.92 ± 1.95</td>
<td>9.1</td>
<td>26 Nov</td>
<td>24.4 ± 5.8</td>
<td>309.4 ± 169</td>
<td>1.53 ± 0.21</td>
</tr>
</tbody>
</table>

**Significant difference between means.

p < 0.01.

p = 0.015.

p = 0.001.

p = 0.02.
3.5. Flower polymorphism

*S. hyacinthoides* flowers exhibit several polymorphic traits. The flowers have two whorls of colored tepals of light–blue color. On the adaxial side of the tepals of some flowers there are marks ranging from violet to green (Fig. 6). Those marks are especially prominent while the flowers are still closed. In addition, some inflorescences have flowers with yellow pollen (Fig. 7). The pollen color becomes visible only after the anthers open. Open anthers with yellow pollen seem to be smaller than the anthers with more frequently occurring violet pollen. The yellow pollen was viable and germinated on sucrose solution (results not shown).

4. Discussion

4.1. *S. hyacinthoides* life cycle

Being a synanthous geophyte, *S. hyacinthoides* begins its annual cycle by resprouting in early autumn after the first rains. The newly emerged leaves result from previous season’s meristematic activity. Simultaneously, the vegetative and reproductive meristems become active (Figs. 2 and 3). Reproductive meristem activity often causes initiation of another meristem which turns into a daughter bulb (Fig. 1). The leaves emerge and elongate continuously during autumn and rainy winter. The flowering occurs in spring. According to Dafni et al. (1981b), such a long period of leaf activity simultaneously with flowering enables allocation of reserves for the next year and for flowering of the current year at the same time, without risking the depletion of the reserves in the bulb.

In the late spring the leaves dry out and in summer the bulb enters a long dormancy, with the meristem becoming inactive. Summer dormancy is critical for plant survival under unfavorable conditions of the dry and hot summer, while the storage materials in the bulb enable fast resprouting in autumn. Numerous geophytes of the Mediterranean region have a similar life cycle (Hyacinthus, *Tulipa*, *Narcissus*; Rees, 1972; Dafni et al., 1981b; Flahmen and Kamenetsky, 2006).

Various stages of geophyte life cycle are known to flexibly react to environmental changes. Indeed, in the Northern Mediterranean, where winters are colder than in Israel, the leaf growth and flowering of *S. hyacinthoides* are shifted, accordingly, to spring and early summer (Tutin et al., 1980). High environmental temperatures prolong dormancy and inhibit growth. High growth temperature (30–C) resulted in severe inhibition of leaf emergence and root extension (Table 2). Understanding the external stimuli for optimal growth and flowering is crucial for effective domestication of the species.
4.2. Flowering

Inflorescence differentiation of *S. hyacinthoides* begins at mid-autumn, around October–November. Our observations suggest that the time from initiation to fully differentiated inflorescence is about two weeks. As temperature drops during winter, the inflorescence gradually elongates inside the bulb, and as they rise again, during spring, the inflorescence emerges from the bulb and anthesis occurs. Thus, a “warm–cold–warm” sequence is required to complete the annual cycle, as for many other Mediterranean geophytes (Le Nard and De Hertogh, 1993; Hanks, 2002; Flaishman and Kamenetsky, 2006).

A temperature of 25 °C seems to be optimal for inflorescence initiation and differentiation in *S. hyacinthoides*. Bulbs subjected to 25 °C showed the earliest onset of initiation, about a month earlier than in 15 and 30 °C, and reached high initiation percentages (Fig. 4A). At 30 °C, bulbs also showed very high percentage of inflorescence initiation, however, differentiation of the inflorescence was arrested at a very early stage (Fig. 4B,C). Therefore, the summer temperature of 30 °C is clearly not suitable for the differentiation step, although it allows induction and initiation. As mentioned, Mediterranean geophytes enter a state of dormancy during the hot dry summer. Thus, the delay of inflorescence development at high temperatures, above 30 °C, prevents differentiation and flowering under unfavorable conditions (Hanks, 2002). But the temperature decrease below 30 °C at the beginning of autumn, allows inflorescence initiation and differentiation. Similar delay of differentiation at 30 °C was also observed for *N. tazetta* (Noy-Porat et al., 2009), and *Crocus sativum* (Molina et al., 2005). After the period of relatively high temperatures needed for initiation and differentiation, a period of lower temperatures is required for inflorescence elongation and maturation (Le Nard and De Hertogh, 1993; Flaishman and Kamenetsky, 2006). Indeed, although 15 °C severely inhibited floral initiation, at the end of the experiment those inflorescences that did develop reached the biggest size (Fig. 4B). A period of autumn–winter cooling was found to be very important for flower differentiation and elongation in numerous geophyte species, of Mediterranean and Irano–Turanian origin, as its absence leads to growth retardation and flowering disorders (Flaishman and Kamenetsky, 2006; Khodorova and Boitel-Conti, 2013). *S. hyacinthoides* requires high temperatures (25–30 °C) for floral initiation, moderate temperatures (<25 °C) for floral differentiation, and low temperatures (~15 °C) for inflorescence elongation and maturation.

*S. hyacinthoides* is able to flower in consecutive years, as the average number of leaves counted between two closest inflorescence Scapes is lower than the number of leaves produced in two growth seasons (Table 1). Its ability to flower almost every year was briefly noted by Dafni et al. (1981b). However, the plants do not flower every year, probably due to unfavorable external conditions. In a “bad year” the plants are either unable to produce enough reserves for flowering, or the temperatures are unfavorable for flower initiation and organogenesis. Under controlled conditions, it will be possible to achieve optimal flowering every year.

4.3. Self-pollination and flower polymorphism

*S. hyacinthoides* is mostly self-incompatible: only about 10% of the flowers are self-fertile, as compared to 90% in cross-pollinated control (Fig. 5). Self-incompatibility in flowering plants prevents self-fertilization and thus both generates and maintains genetic diversity within species. *S. hyacinthoides* is able to reproduce vegetatively by producing daughter bulbs. In nature, *S. hyacinthoides* plants tend to grow in large clones, probably being descendants of the same mother plant. The mature bulbs we dissected had an average of 1.6 daughter bulbs (Table 1), meaning one genetically identical daughter bulb is produced almost every year as compared to several hundred sexually produced seeds. Keeping a genetic diversity has its advantages, and self-incompatibility is a good way to insure genetically diverse offsprings.

For cross-pollination, the plant is dependent on pollinators, thus must invest in flower-pollinator communication. We detected several polymorphic traits in *S. hyacinthoides* flowers: tepal marks (Fig. 6) and pollen color (Fig. 7). In snowdrop (*Gallanthus nivalis* L.) similar green tepal marks were proposed to both attract pollinators, and perform photosynthesis (Aschan and Pfanz, 2006).
Indeed, such tepal marks, visible especially when the flowers are still unopened, might function in communication with pollinators. In \textit{Nigella degenii} similar pollen polymorphism was described, with dark pollen being seemingly more reproductively advantageous (Jorgensen et al., 2000). However, the authors suggested that the yellow pollen might offer an advantage in high pollen competition conditions. Both polymorphic traits of \textit{S. hyacinthoides} flowers might serve as a basis for different agricultural cultivar development.

4.4. Summary

Current research provides a basis for future studies on the florogenesis and ecophysiology in \textit{Scilla}, as well as enabling future fine tuning of cultivation protocols for the flower industry.

Acknowledgments

The research was supported by a grant from the Israel Ministry of Agriculture. The authors thank Yael Ephrat for her invaluable advice on plant anatomy.

References


