

Biology of flowering and insect visitors of *Iris aphylla* L. (Iridaceae)

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Abstract: The paper presents the morphology of the flower of *Iris aphylla* L., which is a protected species in many European countries. The study comprises phenological observations of flowering and assessment of nectar and pollen production, and flowering and fruiting abundance. Additionally, the number of insect visitors on the flowers was monitored. The investigations of specimens originating from 4 populations in central-eastern Poland (Wyżyna Lubelska Upland) were conducted in the Botanical Garden of Maria Curie-Skłodowska University. It was found that morphological features of the analysed flowers, i.e. the 6-angular ovary and the perianth tube, which is 2-fold longer than the ovary, are typical for the taxon *I. aphylla* [= *I. aphylla* var. *typica* L.]. The first flowers developed at the turn of April and May at air temperature of 12–13 °C. The plants produced, on average, 3 flowers per stem, 84.2 flowers per 1 m², and 6.2 capsules per 1 m². The flowers released nectar with a moderate amount of sugar and produced high quantities of pollen. *Mononyctus punctumalbum* Herbst. and bumblebees were found to be the main flower pollinators.

Key words: Flowering phenology and abundance, flower morphology, *Iris aphylla*, nectar, pollen

1. Introduction

The *Iris aphylla* L. (leafless iris) is a rhizomatous perennial plant from the family Iridaceae. Its geographic range covers the steppes of south-west Russia, Kazakhstan, Ukraine, and the Pannonian steppe. Its northern occurrence range reaches Germany, the Czech Republic, Slovakia, Hungary, Poland, and Belarus (Webb and Chater, 1980; Zajac and Zajac, 2001). In Poland, the few natural populations of the leafless iris are located in Wyżyna Małopolska Upland and Wyżyna Lubelska Upland (Kazimierzczakowa and Kucharczyk, 2014). In recent years, the presence of only ten populations has been confirmed. There are three localities in Wyżyna Małopolska Upland: Biała Góra Reserve, Komorów, and Podgrodzie; six in Wyżyna Lubelska Upland: Kazimierz Dolny, Szczecyn, Sobianowice, Zawadówka, Tarnogóra, and Czumów (Franszczak-Być and Dąbrowska, 1993; Czarnecka, 1994; Dąbrowska et al., 1998, Kazimierzczakowa and Kucharczyk, 2014); and one on Załazie Mount: the Biebrzański National Park (Werpachowski and Brzosko, 1998). The marginal character of the *I. aphylla* populations, their spatial isolation, and the isolated 'island' localities (valley slopes, mineral islands surrounded by a peat bog) determine the varying characteristics of these populations (Franszczak-

Być and Dąbrowska, 1993; Czarnecka, 1994; Wróblewska, 2003). Marginal populations naturally tend to become rare and endangered. They are usually described as populations with persistent low abundance, poor flowering, sparse fruiting, evident dominance of vegetative reproduction, or low phenotypic and genotypic variability (Mitka, 1997; Wróblewska, 2003, 2008).

All populations of *I. aphylla* in Wyżyna Lubelska Upland (central-eastern Poland) have been analysed since 1989 for determination of the species biology, phenology, abundance, and density as well as their ecological structure and contribution to the structure of xerothermic grasslands (Franszczak-Być and Dąbrowska, 1993; Czarnecka, 1994; Dąbrowska et al., 1998). Since 1991, integrated ex situ and in situ protection measures have been implemented. These measures include creation of conservation collections at the Botanical Garden of Maria Curie-Skłodowska University in Lublin, strengthening small-sized populations, restoration of historical localities, and improvement of habitat conditions (Dąbrowska et al., 2000; Kwiatkowski et al., 2004).

All leafless iris populations in Wyżyna Lubelska Upland exhibit characteristic traits of a marginal population, i.e. sparse flowering and fruiting and clear dominance of

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vegetative reproduction. The number of generative shoots is highly diverse and varies from zero to several percent in individual sites (Franszczak-Być and Dąbrowska, 1993; Dąbrowska et al., 1998, 2000). This proportion is higher only on slopes with southern exposure in Tarnogóra and Czumów, as every year more than a dozen percent of shoots produce flowers and approximately 15% of them set fruits (Czarnecka, 1994). Simultaneously, it is worth emphasising that the problem with fruit setting has been reported from all *I. aphylla* populations in our country (Wróblewska, 2003; Kazimierczakowa and Kucharczyk, 2014).

Leafless iris flowers are protandrous and require cross-pollination for fertilisation. The complex lipped-throated structure of the iris flower limits insects' access to the nectar (Szafer, 1969). Nectar in this type of flowers is located at a certain depth and is only available to the long-tongued bumblebees and butterflies (Goulson, 1999, 2003; Goulson et al., 2002; Willmer, 2011).

The persistent low frequency of fertilised flowers and the sparse fruiting in the marginal *I. aphylla* populations in Poland have prompted the authors of the present study to (1) undertake thorough observations of phenology, flower morphology, flowering and fruiting, (2) observe insect pollinator visits to the flowers, thus providing practical data on the preferences of the visitor entomofauna, (3) estimate the nectar sugar content, and (4) assess the pollen yield. The investigations of specimens originating from 4 populations in central-eastern Poland (Wyżyna Lubelska Upland) were conducted in the Botanical Garden of Maria Curie-Skłodowska University.

2. Material and methods

2.1. Study species and study site

Iris aphylla is a rhizomatous perennial with a 15 to 40 cm high stem and sabre-shaped falcate leaves reaching the inflorescence apex. The generative shoot is leafless, slightly flattened, and branched below half the height; it produces 1–3 (less frequently 4–5) almost sessile dark purple flowers. The green or purplish bracts are dilated. In Poland, the species blooms in May. The fruit is a triangular capsule containing spherical or oval dark brown seeds (Ascherson and Graebner, 1905; Javorka and Csapody, 1979; Webb and Chater, 1980; Hrouda, 2002; Rothmaler and Jäger, 2007).

The leafless iris grows in well-sunlit habitats on hill slopes or steep valley slopes usually characterised by southern exposure, on limestone or loess substrates (Kazimierczakowa and Kucharczyk, 2014). It occurs most commonly in moderately warm and dry grasslands from the alliance *Cirsio-Brachypodium pinnati* as its typical species (Matuszkiewicz, 2007).

The investigations were carried out in 2016–2018 in the Maria Curie-Skłodowska University Botanical Garden in Lublin, central-eastern Poland (51°14'37.2"N, 22°32'25.3"E; 197 m above sea level). The climate of the Lublin region is characterised by the impact of continental air masses. The vegetation period in Lublin lasts 215 days (Woś, 1999). The average annual temperature (2014–2017) is 10 °C. The average annual total precipitation in Lublin is 553 mm. The meteorological data were obtained from Radawiec station.

The botanical garden is located in the western part of Lublin on the Czechówka River. The leafless iris was cultivated in the southern part of the garden on four 1-m² plots. They were spaced at a distance from 15 to 100 m. The plants grew on three sunlit plots and on one shaded plot located on brown soil formed of loess (26% of fractions < 0.02 mm) characterised by 2.75%–5.37% hummus content and pH 7.76–7.94. The substrate on the plots was enriched with chalk limestone. Plants intended to be analysed were collected in 4 natural habitats from Wyżyna Lubelska Upland, i.e. Kazimierz Dolny, Szczecyn, Tarnogóra, and Zawadówka. Individuals with different origins were found in each plot. They were planted in 2012.

2.2. Floral morphology, flowering phenology, insect-visitors, and abundance of flowering and fruiting

The morphological analysis of floral elements was carried out in the full-bloom phase. The following traits were determined: the colour of flowers from the sunlit and shaded plots, the diameter of the flower, the length and width of petals, the length and the perianth tube, the length and colour of trichomes on outer petals, the length of the pistil, the length and shape of the ovary, the length, width, and colour of the stigma, the length of the stamen, and the length, width, and volume of the anther. The observations of flowering phenology were performed in accordance with the method described by Łukasiewicz (1984). The flowers' life span and development phases, i.e. the appearance of the first flowers (emergence of several first fully opened flowers), the onset of full bloom (approx. 25% of blooming flowers), the end of full bloom (approx. 75% of withered flowers), and the end of blooming (all flowers withered) were investigated. The observations of each flowering phase were carried out every day at 10:00–11:00 Eastern European Time (EET). The abundance of flowering was determined based on the number of all flowers per 1 m² and mean number of flowers per stem. During the full flowering period, the number of insects visiting the flowers was calculated and three groups were distinguished, i.e. the iris weevil *Mononychus punctumalbum* (Herbst, 1784), *Bombus* sp., and others. The number of insects was estimated at 1-h intervals between 7:00 and 19:00. The number of fruits per 1 m² was calculated as well.

2.3. Pollen and nectar.

Using the formula for calculating the volume of a solid of revolution (Birkholc, 2002):

$$V_p = \pi \int_{x_2}^{x_1} [f(x)^2] dx$$

with the following parameters: anther length 8 mm and width 1 mm, the volume of the anther was calculated. The formula for the volume of a sphere:

$$V_{zp} = \frac{4}{3} \pi R^3$$

with the anther radius of 0.097 mm was used for calculation of the volume of a single grain and the number of pollen grains per anther. Pollen yield was determined as the weight of hydrated pollen produced by 10 flowers and converted into 1 m².

Nectar production was assessed using pipette methods (Jabłoński, 2002). Prior to nectar collection, flower buds were randomly selected and protected from insect visitors using tulle isolators (mesh size < 1 mm). The flowers were kept bagged until nectar sampling. Nectar was collected from 2-day flowers, i.e. male-phase flowers. After nectar collection, the flowers were immediately transported to the laboratory. Nectar samples were collected on three dates in 3 replications; single sample contained nectar from 2–3 flowers. The nectar weight was measured with a WPS-36 analytical balance (RADWAG, Radom, Poland). The nectar sugar concentration was measured using an Abbe refractometer (PZO, Warszawa, Poland). The nectar amount and sugar concentration in the nectar were used to calculate the total sugar mass (in mg) produced in nectar per flower.

2.4. Statistical analyses

The results are presented as mean values and standard deviations (SD). The phenological phases, duration of flowering, length of flower life, number of flowers and fruits were subjected to analysis of variance (Stanisz, 2007). Post hoc comparisons of means were tested with the Tukey HSD test. The level of statistical significance for all analyses was set at $\alpha = 0.05$. All analyses were performed using Statistica v. 10.0 (StatSoft, Poland).

3. Results

3.1. Floral morphology, flowering phenology, insect visitors, and abundance of flowering and fruiting

The flowers of the leafless iris have radial symmetry and a diameter of 7.5 ± 1.4 cm. *I. aphylla* specimens growing on the sunlit plots produced purple-red flowers, while those from the shaded plots had purple-navy flowers. The perianth is fused into a 2.5 ± 0.4 cm long tube. The outer petals are obovate (2.6 ± 0.4 cm \times 6.2 ± 0.8 cm), tapered at the base, with white and brown stripes arranged towards

the interior of the flower (Figure 1A). A streak of hairs, i.e. a 'beard' with a length of 3.3 ± 0.4 cm resembling staminodes, is located along the midrib. The hairs are white and blue in the central part of the petal and yellow at the flower base (Figure 1B). The apices of the petals in the outer whorl are strongly curved outward. The inner petals are erect and slightly curved into the flower interior forming a so-called dome (2.7 ± 0.6 cm \times 5.9 ± 0.6 cm); the base of the petals is tapered and has white-brown stripes. The length of the pistil is 3.7 ± 0.2 cm, including the ovary length of 1.3 ± 0.2 cm. The ovary is 6-angular, almost cylindrical, with a diameter of 0.5 ± 0.07 cm. The nectaries are located at the style base. The purple petaliform stigmata end with triangular lobes. Their mean width and length are 1.3 ± 0.09 cm and 3.6 ± 0.09 cm, respectively (Figure 1C). There are large stamens under the stigmata resembling coloured petals (Figure 1D). The 4.5 ± 0.7 cm long stamen has a head with a laterally oriented 0.8 ± 0.1 cm long and 0.1 ± 0.04 cm wide anther. The anther volume V_p was estimated at 16.75 mm³. The leafless iris flowers are protandrous.

In Lublin (central-eastern Poland), the leafless iris began flowering in early spring. The time and length of flowering of the species were associated with the weather conditions prevailing in the preflowering period and during the flowering. The first flowers developed at air temperature of 12–13°C (Figure 2). The onset of flowering exhibited significant variability in the study years ($df = 2$, $F = 67.22$, $P = 0.012$). The difference between the onsets of flowering in the extreme years was 19 days. In 2016 and 2018, which were characterised by higher average daily air temperatures in April and May, *I. aphylla* began flowering on May 3 and 26, respectively, and full bloom was noted in the first 10 days of May. The temperature in spring 2017 was low; hence, the first flowers opened as late as on May 14, and massive bloom was noted already on the 2nd day of flowering (Table 1, Figures 3A and 3B). The length of the flowering period was 12 ± 2.9 days and was significantly different between years ($df = 2$, $F = 24.66$, $P = 0.001$) (Table 2).

The iris flowers bloomed throughout the day. The intensity of the process was enhanced in hot and sunny weather. The first and the last flowers opened at about 8:00 and 18:00 East European Time (EET), respectively. On average, 60%–70% of the daily sum of flowers opened before 15:00. The mean length of blooming of one flower, which was strictly determined by the weather conditions, was 3 days (Table 2). Along the progressing anthesis process, pollen sacs dehisced, and the greatest number of anthers produced pollen simultaneously between 11:00 and 13:00.

Insect visitors appeared as early as about 7:00 and ceased their activity in the afternoon. The maximum



Figure 1. Flower morphology *I. aphylla*: A- labiate-throated type of flowers, B- band of hairs a 'beard' on the outside perianth leaves, C- purple petaliform stigmata, D- stamens in flowering phase, E- mature pollen grains LM, F- pollen grains SEM.

intensity of visits was noted in the morning hours, with a peak between 11:00 and 12:00, when 7–10 insects per 1 m² of the plot were observed, especially in sunlit areas (Figures 4 and 5A). The greatest numbers of insects visiting the leafless iris flowers were represented by the beetle *Mononychus punctumalbum* (80%), different bumblebee species: *B. hortorum* L., *B. lapidaries* L., *B. pascuorum* Scopoli., and *B. terrestris* L., (16%), and others (4%) (Table 3). It was observed that *M. punctumalbum* often chewed oval holes in the petals (Figure 5B) and cut off the anthers (Figure 5C). The mechanical damage shortened the life span of the flowers.

On average, the plants produced 3 flowers per stem. During the three subsequent years of the study, there were, on average, 84.2 flowers per 1 m² (Table 4). Plants growing on the shaded plot produced approximately 30% fewer flowers than the individuals from the sunlit plot. The mean number of seed capsules per 1 m² was 6.2 ± 5.9 , compared with the minimum average of 4.2 ± 2.2 from 2018 and the maximum value of 9.0 ± 11.6 from 2016 (Table 4, Figure 3C).

3.2. Pollen and nectar

The *I. aphylla* flower has 3 stamens. They produce monosulcate heteropolar pollen grains (Figure 1E). The

sulcus is located on the distal side; it is widely open, long, and rounded at the edges. The sulcus surface is smooth. The reticulate exine consists of luminae characterised by varied shapes (spherical, polygonal) and sizes (2–10 μm). Within the luminae, there are circular convex structures. The muri is smooth. The grains are oblato-spheroidal and their size ranges from 74.5 μm to 81.2 μm (Figure 1F). One anther contained on average 4384 pollen grains. The mean weight of hydrated pollen was 0.55 ± 0.07 mg per anther and 16.66 ± 0.10 mg per flower. A mean of 0.14 g of pollen per 1 m² of the plot area was produced by the iris flowers. A single iris flower secreted on average 21.90 mg of nectar with a 16.24% concentration of sugars. The weight of sugars contained in the nectar produced by one flower was 3.38 mg.

4. Discussion

The *Iris aphylla* flowers from the analysed populations in Wyżyna Lubelska Upland (Kazimierz Dolny, Szczecyn, Tarnogóra, Zawadówka) had the purple flowers with a diameter of 7.5 ± 1.4 cm. The perianth was fused into a narrow tube with a length of 2.5 ± 0.4 cm. The elongated lingular outer petals (6.2 ± 0.8 cm long and 2.6 ± 0.4 cm

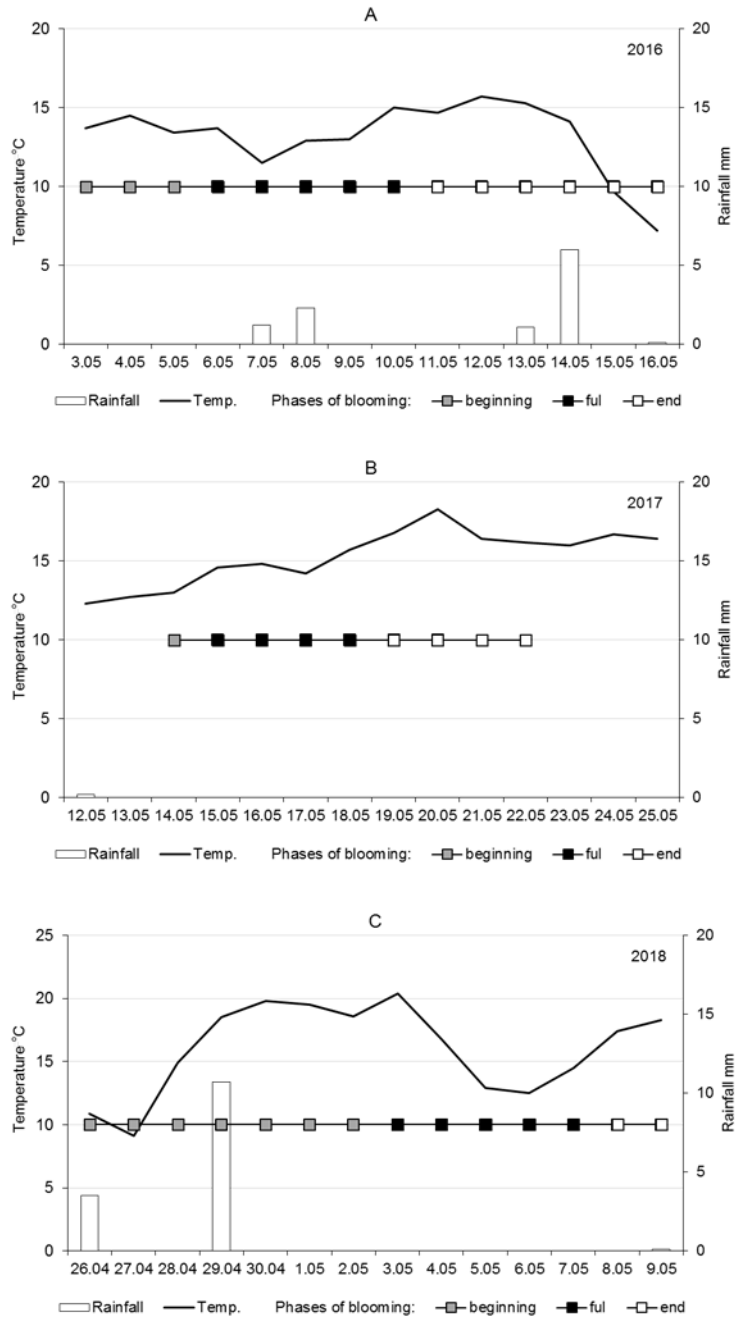


Figure 2. A, B, C – *I. aphylla* flowering in relation to the weather conditions prevailing in 2016–2018 in Lublin, Poland.

wide) were curved outwards. The inner petals (5.9 ± 0.6 cm long and 2.7 ± 0.6 cm wide) were erect and slightly curved towards the interior of the flower. The pistil was 3.7 ± 0.2 cm long, with 1.3 ± 0.2 cm occupied by the ovary. The almost oval 6-angular ovary had a diameter of 0.5 ± 0.07

cm. The morphological features of the analysed flowers, i.e. the 6-angular ovary and the perianth tube, which is 2-fold longer than the ovary, are typical for the taxon *I. aphylla* L. [= *I. aphylla* var. *typica* L.] growing in Poland (Kulczyński, 1919; Webb and Chater, 1980). The morphological features

Table 1. Phenological phases of *I. aphylla* flowering in 2016–2018 in Lublin, Poland. Mean values \pm SD are given. Means within columns with the capital letter (A) do not differ significantly between the years based on Tukey's test at $\alpha = 0.05$.

Year	Phenological phases (days)							
	Beginning of flowering		Beginning of full bloom		End of full bloom		End of flowering	
	Mean \pm SD	V (%)	Mean \pm SD	V (%)	Mean \pm SD	V (%)	Mean \pm SD	V (%)
2016	3.05 \pm 1.5	46.1	6.05 \pm 0.5	8.0	10.05 \pm 0.8	8.2	16.05 \pm 1.7	11.2
2017	14.05 \pm 0.5	3.5	15.05 \pm 1.3	8.2	18.05 \pm 0.5	2.8	22.05 \pm 1.5	6.9
2018	26.04 \pm 0.8	3.1	3.05 \pm 0.5	15.4	7.05 \pm 1.0	14.2	9.05 \pm 1.0	10.9
Mean	4.05	75.2	8.05 ^A	65.2	11.05 ^A	42.4	15.05 ^A	37.1



Figure 3. Phases of plant development: A- flower buds appearing, B- full flowering, C- full fruit ripening.

of the flowers assessed in accordance with the studies by Ascherson and Graebner (1905), Kulczyński (1919), Webb and Chater (1980) are characteristic for the typical form of *I. aphylla* L. [= *I. aphylla* var. *typica*] growing on the steppes in south-west Russia, Kazakhstan, Ukraine, and central Germany. Javorka and Csapody (1979) and Hrouda (2002) distinguished the subspecies *I. aphylla* subsp. *hungarica* (Waldst. & Kit.) Helgi, which occurs on the border of the geographical range of the typical form in Slovakia,

Hungary, Romania, Ukraine, and Moldova. In contrast to the typical form, this subspecies produces smaller flowers with up to 2-cm wide petals and a 3-angular ovary.

Authors of some studies claim that the colour of petals distinguishes the botanical varieties of *I. aphylla*. The present observations suggest that the change in the colour is not associated with the biology of the species but is influenced by environmental conditions. *I. aphylla* specimens growing on the sunlit plots produced purple-

Table 2. Flowering of *I. aphylla* in 2016–2018 in Lublin, Poland. Mean values \pm SD are given. Means within columns with the capital letter (^A) do not differ significantly between the years based on Tukey's test at $\alpha = 0.05$.

Year	Flowering time (days)			
	Duration of flowering		Length of flower life	
	Mean \pm SD	V (%)	Mean \pm SD	V (%)
2016	13.2 \pm 2.6	19.8	3.1 \pm 0.1	4.9
2017	8.5 \pm 1.3	15.2	2.5 \pm 0.6	23.1
2018	13.7 \pm 0.5	3.6	3.2 \pm 0.5	15.4
Mean	11.8 ^A	24.6	2.9 ^A	17.9

red flowers, while those from the shaded plots had purple-navy flowers.

The present study demonstrated that the beginning and length of *I. aphylla* flowering differed substantially between the vegetation seasons. This phenomenon, which is common for many species flowering in early spring (Dąbrowska, 2012; Denisow et al., 2014). In Lublin, the leafless iris flowered in the garden conditions in the first and second 10 days of May. In natural habitats, *I. aphylla* blooms during the flowering period of *Prunus spinosa* L., i.e. in the second and third 10 days of May (central-eastern Poland) (Czarnecka, 1994) as well as the third 10 days of May and the first ten days of June (north-eastern Poland) (Wróblewska, 2003). At this time, the vegetation of most grassland entomophilic species begins.

The leafless iris is a photophilous species, which strongly reacts to changes in habitat conditions, mainly shading. Plants growing in the shade persist in grasslands

in the vegetative stage for many years (Kazmierczakowa and Kucharczyk, 2014). In the plots located in the Garden, there were 84.2 flowers and 6.2 capsules per 1 m² area.

I. aphylla can attract insect pollinators with its colourful perianth, flower size, comfortable landing platform (outer petals), and abundant production of pollen and nectar. In the study period, increased activity of the beetle *Mononychus punctumalbum* was noted. There were 2–8 individuals of the species in one flower at the same time. During feeding, the bodies of the insects were abundantly covered with pollen, which may indicate that they mediate the pollination of *I. aphylla* flowers. Additionally, the iris flowers served as a mating site for these beetles. Besides foraging, this function is highly important for conservation of the biodiversity of insect pollinators as well as preservation and maintenance of various entomophilic plants (Goulson et al., 2002; Sapir et al., 2005). However, the frequency of insect visits was associated with considerable destruction of flowers, as the beetles consumed parts of their perianth and cut off their anthers. It is worth emphasising that intensive *M. punctumalbum* feeding on flowers in natural habitats may lead to damage to the reproductive organs of the flower and limit the generative reproduction of plants, thereby leading to reduction of their population. This issue seems important in light of our earlier findings that the generative reproduction of some European irises, e.g., *I. spuria* L., *I. sibirica* L., and *I. pseudacorus* L., is impaired by *M. punctumalbum* foraging (Skuhrovec et al., 2018). The authors of the work suggest that bumblebees are the best pollinators of throated-type flowers. The reasons for bumblebees' preference for this type of flowers are interesting. Our observations suggest that the morphological traits of *I. aphylla* flowers limit the range of insect visitors more efficiently than the nutritional value of

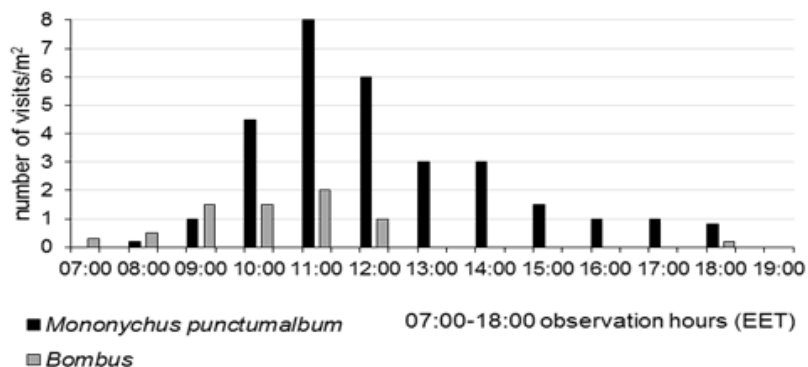


Figure 4. Daily frequency of visits of *I. aphylla* flowers by *M. punctumalbum* and *Bombus* sp. in 2016–2018. Number of insects noted at 1-h intervals.



Figure 5. *Mononychus punctumalbum* visiting flowers *I. aphylla* and signs of feeding: A- flower visited by *M. punctumalbum*, B- gnawed holes in perianth blades, C- bitten stamen.

Table 3. Percentage proportion of the insect-visitor groups on *I. aphylla* flowers.

Year	<i>Mononychus punctumalbum</i>	<i>Bombus</i> sp.	Other
2016	78.1	16.8	5.1
2017	82.6	16.5	0.9
2018	79.3	14.7	6
Mean	80	16	4

nectar. Iris flowers are closed due to the fusion of the outer petal and the lobular stigma. To collect nectar, insects must

open these elements of the flower. Additionally, the nectar in the flower is located on the bottom of the 2.5 cm long perianth tube. It is known that the characteristics of nectar determine the spectrum of insects visiting various plant species (Galletto and Bernardello, 2004). As demonstrated by various authors, bumblebees most often collect nectar with a 25%–40% concentration of sugars, whereas honeybees prefer a sugar concentration range of 50%–65% (Nicolson and Thornburg, 2007). Iridaceae are a highly diverse family in terms of nectar production. Flowers in the genus *Iris* tend to release secretion less concentrated nectar (Rudall et al., 2003). The concentration of sugar

Table 4. Abundance of flowering and fruiting in *I. aphylla* in 2016–2018 in Lublin, Poland. Mean values \pm SD are given. Means within columns with the capital letter (A) do not differ significantly between the years based on Tukey's test at $\alpha = 0.05$.

Year	Abundance of blooming				Abundance of fruiting	
	Number of flowers per stem		Number of flowers per 1 m ²		Number of fruits per 1 m ²	
	Mean \pm SD	V (%)	Mean \pm SD	V (%)	Mean \pm SD	V (%)
2016	3.5 \pm 0.4	12.3	95.2 \pm 16.0	16.8	9.0 \pm 11.6	129.3
2017	3.5 \pm 0.4	12.5	75.2 \pm 31.5	81.8	5.5 \pm 3.9	70.4
2018	3.6 \pm 0.3	8.3	82.0 \pm 10.9	13.3	4.2 \pm 2.2	52.2
Mean	3.5 ^A	11.0	84.2 ^A	25.1	6.2 ^A	84.0

may be associated with the nectary traits and type of sucrose transport via the nectary tissue (Nicolson and Thornburg, 2007). Differences in the concentration of sugar in nectar can also be caused by changing weather conditions (air humidity and temperature) (Denisow et al., 2014). In the present study, the leafless iris nectar with a lower sugar concentration (approx. 16%) was collected mainly by bumblebees (*B. hortorum*, *B. lapidarius*, *B. pascuorum*, *B. terrestris*). In general, the relationship of the phenotypic floral traits and the floral-reward traits with the frequency of insect visits is a common phenomenon of interdependence (Goulson et al., 2002; Willmer, 2011).

A majority of anthers in the leafless iris flowers dehisced at 11:00–13:00. This feature is common to many spring plant species (Dąbrowska, 2012). The mechanism of anther dehiscence in the middle of the day is associated with the weather conditions, e.g. a decline in relative humidity and a rise in air temperature. Such conditions increase water loss from anther tissues and facilitate pollen release (Pacini, 2000). The time of pollen release in spring plants corresponds to the daily pattern of insects' activity. A single *I. aphylla* flower produced over 13,000 large pollen grains, which were an abundant source of protein for beetles, bumblebees, and bees. In terms of taxonomic classification, *I. aphylla* pollen grains represent the series *Elatae* (Mitić et al., 2013). Hydrated pollen grains of this type are relatively large (up to 100 μ m) with an oblatospheroidal shape and a smooth sulcus extending to half of the grain circumference. They have a heterobrochate-reticulate exine with large luminae (up to 10 μ m in diameter). The series *Elatae* also comprises other European

irises, i.e. *I. croatica* L., *I. germanica* L., *I. pallida* Lam., *I. pallida* subsp. *Cengialti* Lam., *I. pallida* subsp. *illyrica*, Lam., *I. pseudopallida* Trin., *I. reichenbachii* Heuffel., and *I. variegata* L.. Given the morphological traits of the pollen grains in these taxa, it can be concluded that a majority of garden varieties emerging via crossing the taxa are a source of valuable food for insects.

Finally, *Iris aphylla* produced flowers with typical morphological features of the taxon form *I. aphylla* L. [= *I. aphylla* var. *typica*] i.e. a 6-angular ovary and a 2-fold longer perianth tube.

I. aphylla begins growing in early spring and, depending on weather conditions, flowers between the end of April and mid-May. The plants produced, on average, 3 flowers per stem, 84.2 flowers per 1 m², and 6.2 capsules per 1 m².

The attractive and fragrant *I. aphylla* flowers attracted insects, mainly the *M. punctumalbum* beetle and *Bombus* sp. Pollen (16.66 mg per flower) and nectar (21.90 mg) with a sugar concentration of 16.24% were the main rewards for insect visitors.

The beetles visiting the flowers did not collect nectar but consumed pollen together with anthers. They often damaged the flowers but concurrently pollinated the flowers while they were feeding and flying from one flower to another. Intensive foraging by the beetle can result in the low generative reproduction of this species.

Bumblebees were the only Apidae representatives that were able to overcome the morphological barriers of the flower. They parted fused petals and lobular stigmata and reached the nectar deposited on the bottom of the relatively long perianth tube.

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