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ORIGINAL RESEARCH PAPER

Nectar and pollen production in *Arabis procurrens* Waldst. & Kit. and *Iberis sempervirens* L. (Brassicaceae)

Monika Strzałkowska-Abramek[1]*, Karolina Tymoszuk[2], Jacek Jachuła[1,2], Małgorzata Bożek[1]

[1] Department of Botany, University of Life Sciences in Lublin, Akademicka 15, 20-950 Lublin, Poland
[2] Biological Students Research Group, Faculty of Horticulture and Landscape Architecture, University of Life Sciences in Lublin, Akademicka 15, 20-950 Lublin, Poland

* Corresponding author. Email: monika.strzalkowska@up.lublin.pl

Abstract

Ecological environment in urban areas is specific in many aspects. There are evidences that ornamental plants cultivated in local urban gardens may help in conservation of pollinators. In this study, the flowering pattern, the abundance of flowering, nectar and pollen production as well as insect visitation in *Arabis procurrens* Waldst. & Kit. and *Iberis sempervirens* L. were investigated. The species were grown in the UMCS Botanical Garden in Lublin, southeastern Poland. *Arabis procurrens* bloomed from the middle of April until middle of May and *I. sempervirens* from the end of April until middle of June. In both species, most flowers opened in the morning hours (40–45% of total were opened by 8:00 h GMT + 2 h). The average sugar yield of *A. procurrens* was ca. 53% lower compared to *I. sempervirens* (mean = 1.08 g/m^2 and 2.32 g/m^2, respectively). In both species, considerable differences in the amount of produced sugars were noted between years. The mass of pollen produced in the flowers of *A. procurrens* was approx. 35% lower compared to that of *I. sempervirens* (mean = 0.06 mg and 0.09 mg per flower, respectively). Pollen produced per unit area was correlated with the number of flowers. On average, the species produced 1.46 g (*A. procurrens*) and 2.54 g (*I. sempervirens*) of pollen per 1 m^2. The flowers of *A. procurrens* attracted mainly dipterans (56.3% of total visitors), while *I. sempervirens* lured chiefly solitary bees (47.4% of total visitors), however in both cases, honeybees, bumblebees and lepidopterans were also recorded. The *A. procurrens* and *I. sempervirens* due to flowering in early spring period may be promoted for use in small gardens (rock or pot gardens) for both aesthetic value and as plants that support insect visitors in nectar and pollen rewards.

Keywords

diurnal pattern of blooming; nectar yield; pollen yield; insect visitors

Introduction

Nectar and pollen are floral primary attractants [1]. They provide the complete diet for both the adults and the larvae of insect pollinators [2]. Nowadays, due to anthropogenic pressure (intensification in agriculture, herbicide use) food plants are frequently destroyed [3–5]. Disappearance of flower-rich patches from the landscape results in decrease of bees and other pollinator populations in many parts of the world [2]. To counteract the pollinator decline, the restoration and promotion of flowering plants that ensure food resources is necessary both in urban [6] and agricultural areas [7]. Cultivation of nectar and pollen plants, including ornamentals, may help pollinators [8–12]. Therefore, the evaluation of nectar and pollen reward is one of the ways to select ornamentals that are beneficial to bees [13]. Thanks to flower morphology, Brassicaceae species are recognized attractive to insects [1,14,15].

Ornamental gardens are usually designed for their aesthetic pleasure. Plethora of foliage plants, ornamental grasses, or double-flower plants which are useless for insect visitors are grown [11,16]. Currently, rock gardens are very popular and many mass-flowering species are recommended [17]. However, the value of floral reward may differ considerably between species or even cultivars [18].

The aim of this study was to evaluate the value of floral reward in two Brassicaceae species (*Arabis procurrens* Waldst. & Kit. and *Iberis sempervirens* L.), propagated for modern rock garden design due to their slow-growing properties, delicacy, the attractiveness of flowers and leaves [17]. More specific goals were to examine (***i***) blooming biology, (***ii***) nectar secretion, (***iii***) pollen production, and (***iv***) the spectrum of insect visitors.

Material and methods

Study site

The observations were conducted in the years 2014–2015. The species were grown fully exposed to the sun in the alpine section of the UMCS Botanical Garden in Lublin, Poland (51°16′ N, 22°30′ E).

Study species

Two perennial species, *Arabis procurrens* Waldst. & Kit. and *Iberis sempervirens* L., were selected for the study. The species differ in terms of their origin: *A. procurrens* is native to Europe and occurs in Carpathian and Balkan mountains, whereas *I. sempervirens* is widespread in South Europe, North Africa and West Asia [17].

Flowering and insect visitors activity

The methods described by Denisow [19,20] were applied. We established the onset and length of blooming, as well as the pheno-phases duration. The initiation of blooming was when 10% of flowers started to bloom, the full blooming was when 70% started to bloom, and the termination when 80% fell off. In 2014, we observed the diurnal pattern of blooming. The observations were conducted from 6:00 till 19:00 (GMT + 2 h), and newly opened flowers ($n = 5$ inflorescences per species) were counted in one-hour interval. Together with the blooming observations, insect visits were noted at the full bloom phase (3 days, $n = 3$ plots of 1 m^2). During each census of observation, we recorded the total number of visiting insects and the type of gathered forage (nectar vs. pollen). The number of flowers per raceme ($n = 24$–30), and the number of inflorescences on random circular areas 0.1 m^2 (36.7 cm in diameter) were established. The data were converted to the number of flowers per 1 m^2 of the surface, and were used to estimate the total nectar and pollen yield.

Nectar secretion

We examined the secretion of nectar using the pipette method [21]. Prior to nectar collection, the inflorescences were isolated from insect visitors with the tulle isolators. Nectar was collected in 2–3 replications during blooming period. In each replication 3–5 samples were collected, a single sample contained nectar from 8–11 flowers. Total sugar concentration was measured with Abbe refractometer. Nectar amount and sugar concentrations were used to calculate the total sugar mass in each sample. Relevant calculation allowed to determine the amount of sugars produced per ten flowers (in mg) and per 1 m^2 (in g).

Pollen production

The mass of pollen was examined in the full blooming phase. Mature but unopened anthers (n = 60) were collected in tarred glass containers (n = 4) [20]. Subsequently, the glass containers with anthers were placed into a dryer (ELCON CL 65) at ca. 33°C. The pollen was rinsed from anthers with ether and with ethanol (70%). The pollen production was calculated per ten flowers (in mg) and per 1 m^2 (in g).

Weather conditions

According to the Institute of Meteorology and Water Management, long-term average temperature/precipitation data for Lublin were as follows: March 1.1°C / 26.3 mm; April 7.5°C / 40.2 mm; May 13.0°C / 57.7 mm; June 16.2°C / 65.8 mm. The spring of 2014 was an early one, with temperatures 3.5°C higher than long-term average in March and 2.5°C in April. In addition, the heavy drought was recorded, i.e., in April the rainfall was 27.2 mm, and periodic considerable drops of temperature were noted (−10°C). On the contrary, in May heavy rainfalls (2-fold higher than long-term norm) were noted. In the study region, at the end of May and the beginning of June 28–33°C air temperature were recorded. The spring of 2015 was also an early one, with mild temperatures in March and April, but a spring was rather dry, i.e., in April the rainfall was 19.5 mm, and June 11.3 mm.

Data analysis

Data in tables are presented as means with *SD*. The one-way ANOVA was applied to test the significance of differences [22]. Post hoc comparison of means was tested by the Tukey's test. The level of statistical significance for all analyses was at p = 0.05. All analyses were performed using Statistica ver. 6.0 (StatSoft Poland, Cracow).

Results

Under the climatic conditions of eastern Poland, during the study seasons, the blooming of studied perennials occurred in April/May (Fig. 1). The blooming of *Arabis procurrens* initiated 10–14 days before that of *Iberis sempervirens*. The length of flowering period differed between species: it ranged from 28 to 30 days for *A. procurrens* (mean = 29.0) and was three weeks shorter than flowering length of *I. sempervirens*, which ranged 49–59 days (mean = 54.0). The length of flowering in 2014 differed significantly from that in 2015 only for *I. sempervirens*.

Flowers of *A. procurrens* and *I. sempervirens* are aggregated in racemes and commenced blooming acropetally, with an early diurnal flower-opening pattern (Fig. 2). About 40–45% of daily installment was observed at 7:00–8:00 h (GMT + 2 h), then a steady progress of the process was noticed, and approximately 3–10% of flowers opened every 1–2 hours till 17:00–18:00 h, when the day's anthesis terminated.

The species effect was found for the number of flowers per inflorescence ($F_{1,75}$ = 36.61, p = 0.004) and for the number of shoots per 1 m^2 ($F_{1,7}$ = 16.34, p = 0.001) but not for the number of flowers per 1 m^2 ($F_{1,7}$ = 12.45, p = 0.211; Tab. 1). The single raceme of *I. sempervirens* developed almost 4-fold more flowers (mean = 121.1) than raceme of *A. procurrens* (mean = 33.4). The significant year-to-year differences in the number of flowers per inflorescence ($F_{1,29}$ = 7.39, p = 0.012 – *A. procurrens*; $F_{1,29}$ = 4.35, p = 0.044 – *I. sempervirens*), in the number of shoots per 1 m^2 ($F_{1,7}$ = 9.23, p = 0.012 – *A. procurrens*; $F_{1,7}$ = 5.38, p = 0.034 – *I. sempervirens*), and in the number of flowers per 1 m^2 ($F_{1,7}$ = 7.9 , p = 0.012 – *A. procurrens*; $F_{1,7}$ = 7.9, p < 0.001 – *I. sempervirens*) were noted. The intra-species disparities in the abundance of blooming per unit area between years were ca. 2-fold.

The amount of secreted nectar ($F_{1,36}$ = 34.05, p = 0.021) varied significantly between species, however, no species effect was found for the concentration of sugars in

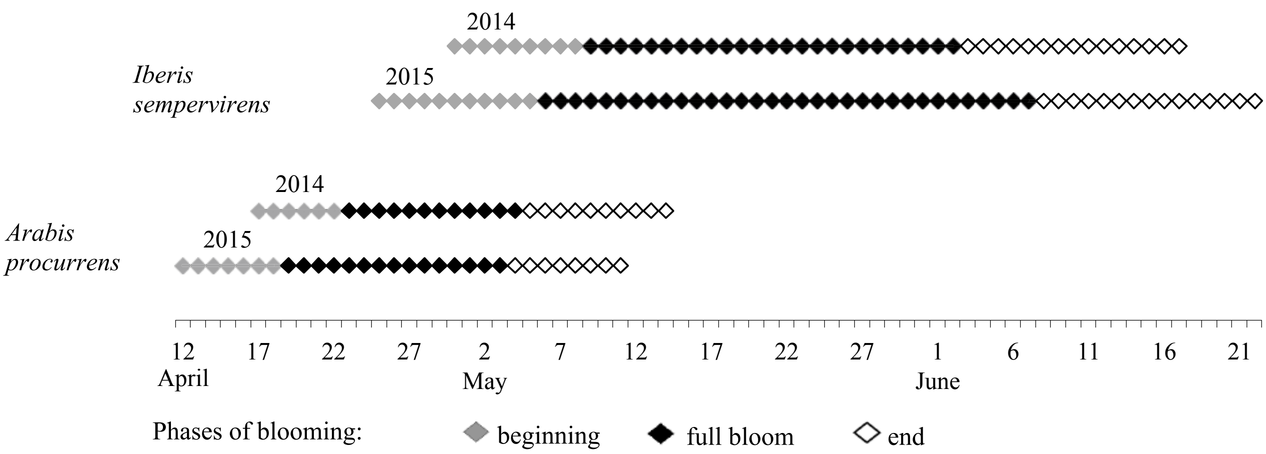

Fig. 1 The time and period of blooming of *Arabis procurrens* and *Iberis sempervirens* in Lublin in the years 2014–2015.

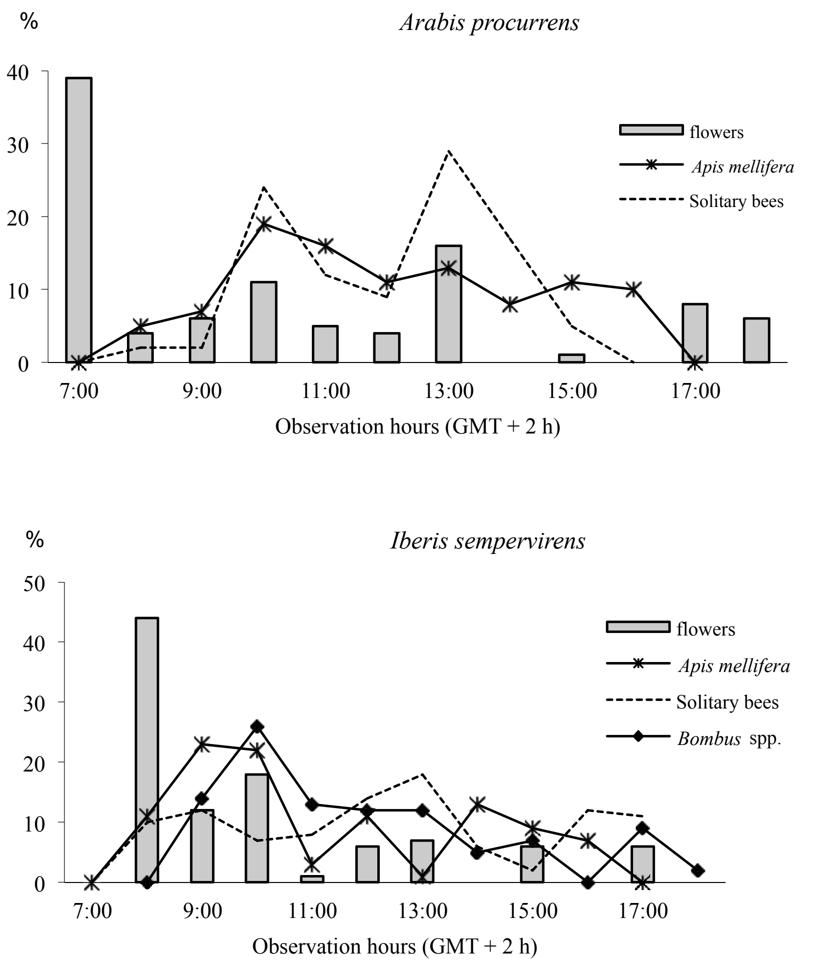

Fig. 2 Diurnal pattern of flowering of *Arabis procurrens* and *Iberis sempervirens* expressed as the number of flowers opened at 1-h intervals in relation to the total opened during the day, and the diurnal activity of Apidae visitors observed in SE Poland (means calculated from 2014–2015).

nectar ($F_{1,36} = 16.62$, $p = 0.071$; Tab. 2). The flowers of *I. sempervirens* secreted 2-fold more nectar than those of *A. procurrens* (mean = 0.16 mg per flower vs. 0.08 mg per flower, respectively). The species effect was found for the total sugar mass produced in flowers ($F_{1,36} = 12.90$, $p = 0.037$). The average sugar yield of *A. procurrens* was approx. 53% lower compared to *I. sempervirens* (mean = 1.08 g/m^2 and 2.36 g/m^2, respectively). For both species, considerable differences were noted between years: 2-fold disparities were recorded in the case of *A. procurrens* and 3.5-fold in the case of *I. sempervirens* (Fig. 3).

The species effect for the size of anthers (expressed as anther dry weight) was established ($F_{1,15} = 11.32$, $p = 0.046$), and for the mass of pollen produced in flowers ($F_{1,15} = 9.92$, $p = 0.042$); 1.5-fold more pollen was produced in the flowers of *I. sempervirens* compared to *A. procurrens* (Tab. 3). However, both species had different pattern of pollen production. Namely, the pollen output of *A. procurrens* was relatively stable between years ($F_{1,6} = 3.92$, $p = 0.059$), whereas the flowers of *I. sempervirens* produced 43% more pollen in 2015 than they did in 2014 ($F_{1,6} = 18.25$, $p < 0.007$).

The flowers of *A. procurrens* attracted mainly dipterans (56.3% of total visitors), while *I. sempervirens* lured chiefly solitary bees (47.4% of total visitors), however, less numerous visits of insects from other surveyed groups (i.e., honey bees, bumblebees and lepidopterans) were also recorded in both species (Fig. 4).

Tab. 1 The total abundance of blooming of *Arabis procurrens* and *Iberis sempervirens* in the years 2014–2015, observed in SE Poland. Mean values and standard deviation (*SD*) are given.

Species	Year	Number of flowers per raceme			Number of shoots per 1 m^2			Number of flowers per 1 m^2 (thous.)		
		min.–max.	mean	±*SD*	min.–max.	mean	±*SD*	min.–max.	mean	±*SD*
Arabis procurrens	2014	24–69	29.8_a	24.5	395–684	584.2_a	95.8	14.0–40.3	17.4_a	3.6
	2015	14–73	37.0_b	14.4	725–864	790.0_b	70.0	11.1–57.7	29.2_b	8.2
	mean		33.4_A			687.1_A			23.3_A	
Iberis sempervirens	2014	37–189	103.8_a	23.1	137–156	146.3_a	19.5	5.4–27.7	15.2_a	4.1
	2015	55–287	138.3_b	71.4	260–420	286.6_b	32.8	15.8–82.3	39.8_b	3.8
	mean		121.1_B			216.5_B			27.5_A	

Means within columns with the same small letters are not significantly different between years, and followed by the same capital letters are not significantly different between species, according to Tukey's test at $p = 0.05$.

Tab. 2 Nectar amount and concentration and amount of sugars produced by *Arabis procurrens* and *Iberis sempervirens* in the years 2014–2015, observed in SE Poland. Mean values and standard deviation (±*SD*) are given.

Species	Year	No. of examined flowers	Nectar amount per flower (mg)			Concentration of sugars in nectar (%)			Sugar amount per flower (mg)		
			min.–max.	mean	±*SD*	min.–max.	mean	±*SD*	min.–max.	mean	±*SD*
Arabis procurrens	2014	110	0.06–0.10	0.07_a	0.02	49–68	52.3_a	2.9	0.03–0.07	0.04_a	0.01
	2015	112	0.09–0.14	0.10_b	0.04	45–59	49.8_b	4.1	0.04–0.08	0.05_b	0.01
	mean			0.08_A			51.1_A			0.05_A	0.02
Iberis sempervirens	2014	120	0.07–0.16	0.14_a	0.08	43–64	49.1_a	4.3	0.03–0.08	0.07_a	0.03
	2015	125	0.10–0.24	0.17_b	0.05	38–67	53.2_b	3.6	0.04–0.16	0.09_b	0.02
	mean			0.16_B			51.2_A			0.08_B	0.03

Means within columns with the same small letters are not significantly different between years, and followed by the same capital letters are not significantly different between species, according to Tukey's test at $p = 0.05$.

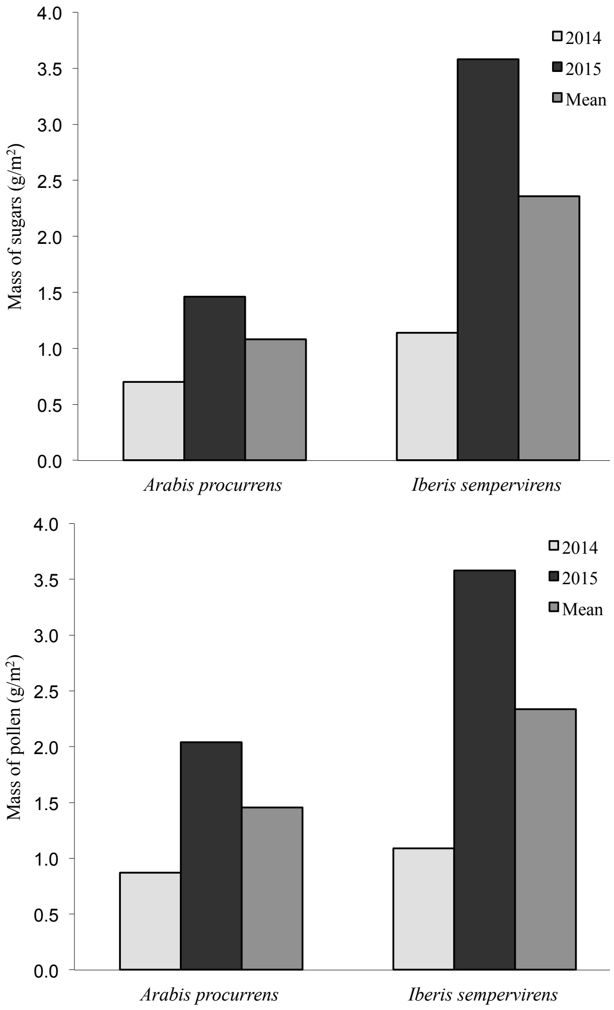

Fig. 3 Sugar and pollen yield of *A. procurrens* and *I. sempervirens* in the years 2014–2015 (SE Poland).

Discussion

The blooming period of *Arabis procurrens* and *Iberis sempervirens* established in our study is similar to that reported for Poland by Marcinkowski [23]. In accordance to the study of Denisow [18], we noted that flowering of *A. procurrens* began about 10–14 days earlier compared to *I. sempervirens*. Repeatable sequences of flowering indicate that studied species, when arranged together, can support pollinators from the mid-April until the mid-June. The length of blooming was variable between seasons only for *I. sempervirens*. In 2014, the shorter blooming period resulted from the air temperatures that exceeded the long-term mean accompanied by shortages of rainfalls. In the study region, at the end of May and the beginning of June, the 28–33°C air temperature was recorded. The effects of high air temperatures and drought have been reported to accelerate the flower-life span and consequently shorten the period of blooming [9,10,24].

Our survey documented early pattern of flower opening during the day for both studied species, which is congruent with previous reports on blooming of brassicaceans, e.g., wild-growing *Berteroa incana*, *Bunias orientalis* [15], *Sisymbrium loeselii* [25], or ornamental *Aubrieta* ×*hybrida* [18]. In general, Brassicaceae taxa open the majority of flowers early during the day (40–70% are opened before 8:00 h GMT + 2 h).

The abundance of blooming differed between species, as well as intra-species year-to-year differences were noted. *Arabis procurrens* and *I. sempervirens* were classified among abundantly blooming perennials, as they had more than 60 000 flowers per 1 m² [18]. In our survey, both species produced approx. 50% less flowers. Multiple factors may explain the disparities. For perennial plants, the plant age may significantly affect the number of flowers per inflorescence, per individual and per unit area [26,27]. As shown for many perennials, flower production in such

Tab. 3 The dry mass of anthers, the mass of produced pollen, total pollen yield and pollen viability in *Arabis procurrens* and *Iberis sempervirens* in the years 2014–2015, observed in SE Poland. Mean values and standard deviation (*SD*) are given.

Species	Year	Mass of 100 anthers with pollen (mg)		H_2O (%)	Mass of pollen per 10 flowers (mg)		
		fresh	dry		min.–max.	mean	±*SD*
Arabis procurrens	2014	20.9	10.7_a	48.8_a	0.3–0.8	0.5_a	0.1
	2015	24.8	12.4_b	50.0_a	0.4–1.0	0.7_a	0.2
	mean	22.4	11.6_A	48.1_A		0.6_A	
Iberis sempervirens	2014	16.9	6.3_a	62.7_a	0.5–1.0	0.7_a	0.2
	2015	20.4	7.2_b	64.7_a	0.7–1.4	1.0_b	0.4
	mean	18.7	6.8_B	63.7_B		0.9_B	

Means within columns with the same small letters are not significantly different between years, and followed by the same capital letters are not significantly different between species, according to Tukey's test at $p = 0.05$.

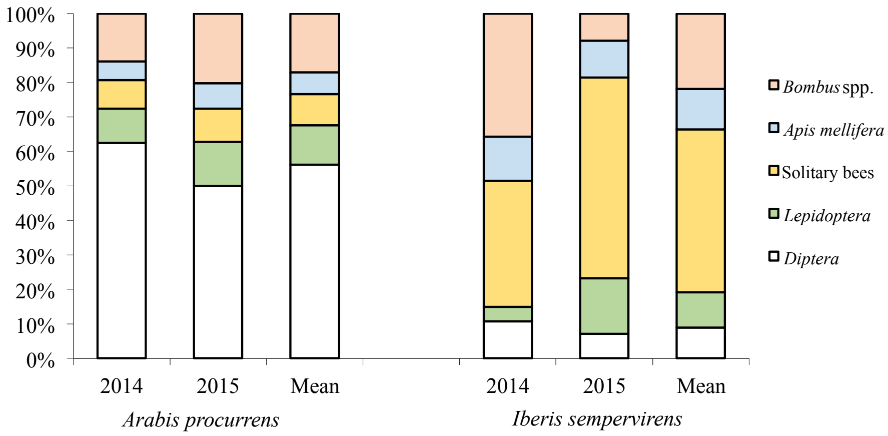

Fig. 4 The composition of insect visitors of *A. procurrens* and *I. sempervirens* in the years 2014–2015 (SE Poland).

plants is partly supported by carbohydrate reserves in the underground rootstock tissue [28].

Moreover, in many plants the abiotic conditions experienced prior or during flowering have been described to influence the bud formation and the number of flowers [24]. In plants growing in mountains conditions (i.e., generally similar to that of alpine gardens) an earlier snowmelt may result in greater exposure to freezing conditions and can cause the flowers damage [1]. In April of 2014, considerable drops of temperatures were noted (−10°C). Year-to-year disparities in abundance of flowering could also be caused by a change in the availability of resources between growing seasons [27,28].

Flowers of *A. procurrens* and *I. sempervirens* are comparable in structure with that of many other brassicacean species; they possess a typical cross-like arrangement of petals, tetradynamous stamens and a single superior pistil [14]. In the studied species, petals persisted only 2–3 days in open flowers of *A. procurrens* and 5–7 days in *I. sempervirens*, which is consistent with the findings of Denisow [18], who reported longer life-span for *I. sempervirens* compared to other brassicacean spring ornamentals. The flower lifespan is species-specific [9,29]. Flower longevity is also under control of various exogenous factors, e.g., weather conditions, especially air temperature and humidity [19,30]. The flower life-span may impact on the plant-pollinator relationship as it determines the rate of floral reward availability [29]. However, the other traits, e.g., color, size, symmetry, shape as well as nectar and pollen quality impact on the plant-pollinator interaction [1]. Flowers of *A. procurrens* and *I. sempervirens* appear to be entomophilous because of their mass flowering and presence of floral reward (pollen and nectar), which is exposed and easily accessible for different groups of insects. These are key features of insect-pollinated plants [1,20]. Despite similar flower morphology, the composition of floral visitors differed between species studied here. Various factors are expected to influence this disparity. Apart from flower morphology, nectar and pollen characteristics impact on insect visitors to flowers, for example – the type of carbohydrates or other compounds, i.e., lipids, phenols, alkaloids, or organic acids [31], an amount of sugars produced per unit area [9], absence/presence of starch or proteins in pollen [20]. These nectar and pollen traits are highly species-specific [1,6,31].

Our study species tend to secrete medium to high concentrated nectar (38–68%), which was in the range reported for other brassicaceans ([1] and references therein). Variable nectar concentration is a very common phenomenon and primarily it could be related to nectary characteristics and type of sucrose transport across the nectary tissue [31]. Moreover, it is usually associated with changeable weather parameters [9], therefore variable air temperature and humidity as well as precipitation may explain the year-to-year differences in nectar sugar concentration noted in our experimental species.

The quantity of nectar sugar and pollen are important factors to consider during the selection of plants for bee-friendly gardens, as the lack of floral resources is indicated among the reasons of pollinators decline [2–4]. Total nectar production in *I. sempervirens* and *A. procurrens* was about 10 and 15 lower compared to values obtained for flowers of cultivated brassicaceans, i.e., spring rapeseed [32] or winter rape [33] as well as wild *Sisymbrium loeselii* [26]. Studied ornamentals differed considerably in nectar production between study years. Nectar production relies on current photosynthesis [31] and it is possible, that differences in weather patterns between seasons impacted on photosynthesis efficiency and affected nectar production differently.

Overall total sugar yield of species studied was low (only 10.8 g per 10 m^2 – *A. procurrens*, 23.2 g per 10 m^2 – *I. sempervirens*), which indicates that the investigated species should be treated as poor sugar yielding plants. Pollen production that amounted 14.6–25.4 g per 10 m^2, was 2–3-fold lower than that recorded earlier for the same species by Denisow [18], and 3–6-fold lower than values obtained for other brassicaceans, e.g., *Arabis caucasica*, *Aubrieta ×hybrida* or similar to that estimated for *Alyssum saxatile* [18]. The differences in pollen production result usually from the mass of pollen produced in anthers, which relate to anther size as well as weather conditions [20]. For example, during the drought even empty anthers occur [18,24]. In addition, the abundance of blooming affects the amount of available food resources [6].

In conclusion, the amount of produced nectar sugars and pollen was relatively low, however, the flowers of *A. procurrens* and *I. sempervirens* are visited by insects from different taxonomic groups, not only Apoidea. This indicates that these plant species are vital in the maintenance of general insect biodiversity and should be popularized for both their aesthetic value and as plants that support the forage of insect visitors during early spring period.

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Produkcja pyłku i nektaru w kwiatach *Arabis procurrens* Waldst. & Kit. i *Iberis sempervirens* L. (Brassicaceae)

Streszczenie

Przeprowadzone badania dotyczyły dynamiki i obfitości kwitnienia, wydzielania nektaru, produkcji pyłku oraz składu entomofauny dwóch gatunków: *Arabis procurrens* Waldst. & Kit. i *Iberis sempervirens* L. uprawianych w Ogrodzie Botanicznym UMCS w Lublinie. *Arabis procurrens* kwitł od połowy kwietnia do połowy maja, natomiast *I. sempervirens* od końca kwietnia do połowy czerwca. U obu gatunków większość kwiatów (40–45%) rozkwitała wczesnym rankiem, tj. ok. 8:00 (GMT + 2 h). Średnia masa cukrów (w przeliczeniu na jednostkę powierzchni) *A. procurrens* była o ok. 53% mniejsza niż w przypadku *I. sempervirens* (odpowiednio 1.08 g/m^2 i 2.32 g/m^2). Stwierdzono istotne różnice w masie produkowanych cukrów pomiędzy latami

badań dla każdego z badanych gatunków. Średnia masa pyłku produkowanego przez kwiaty *A. procurrens* była o ok. 35% mniejsza niż masa pyłku *I. sempervirens* (odpowiednio 0.06 mg i 0.09 mg). Wydajność pyłkowa, wynosząca 1.46 g/m^2 dla *A. procurrens* i 2.54 g/m^2 dla *I. sempervirens*, skorelowana była z liczbą kwiatów. Kwiaty *A. procurrens* oblatywane były głównie przez muchówki (56.3% liczby owadów), a kwiaty *I. sempervirens*- przez pszczoły samotnice (47.4%). Odnotowano również wizyty pszczoły miodnej, trzmieli i motyli. Badane gatunki roślin, ze względu na wczesną porę zakwitania, mogą stanowić uzupełnienie taśmy pokarmowej owadów zapylających.

