

FLOWERING ABUNDANCE AND POLLEN PRODUCTIVITY OF *Ligularia clivorum* Maxim. AND *Ligularia przewalskii* Maxim.

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Abstract. *Ligularia* genus includes species cultivated in house gardens and urban parks as an attractive ornamental plants. During flowering period their flower heads may provide pollen food for pollinators. In the period 2004–2008, the present study investigated the abundance of flowering and pollen production of two *Ligularia* species as a source of pollen for insects. Under the conditions of Lublin, the onset of flowering of *L. clivorum* occurs in the second half of July, whereas *L. przewalskii* starts to bloom already in the last week of June. Depending on species and weather conditions, the period of flowering of *L. clivorum* lasted 48–67 days, while in the case of *L. przewalskii* from 29 to 38 days. In both taxa, female ray florets and bisexual disc florets are found in capitulum inflorescences. The weight of pollen produced by 10 *L. clivorum* flowers was almost twice higher than that in *L. przewalskii*. Pollen production per unit area for these taxa was 6.92 g·m⁻² and 1.53 g·m⁻², respectively.

Key words: Asteraceae, flowering, pollen production

INTRODUCTION

In the flora of Poland, the Asteraceae family is represented by numerous species of both crop plants and plants that occur in natural communities. During the period of flowering, their inflorescences are a source of pollen for honey bees and other pollinating insects [Warakomska 1972; Käpylä and Niemelä 1979; Rudnianskaja 1981; Wróblewska 1995, 1997; Denisow 2011]. Insects interest in collecting this material has been confirmed by microscopic analysis of pollen gathered in the form of pollen loads and bee bread by insects from various systematic groups [Warakomska 1999; Wróblewska 2002; Teper 2004; Wróblewska et al. 2010; Denisow 2011].

Among representatives of this family, ornamental species play an important role, since their flowering lasts from early spring until as late as the first autumn frosts. In

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this group of plants, the *Ligularia* genus deserves special attention; it is represented by more than 100 species that are present mainly in Asia (including more than 80 species in China) where they frequently occur in natural habitats or as weeds [Liu et al. 1994, 2003; Marcinkowski 2002; Rui-Jun et al. 2005]. In the flora of southern Poland, Siberian ligularia (*L. sibirica*), which is a critically endangered, fully protected species, can be found in peat bogs and thickets [Mirek and Piękoś-Mirkowa 2006]. Due to their decorative inflorescences and leaves, different *Ligularia* species are planted more and more frequently in parks and home gardens as well as in flower beds.

The aim of the present study was to investigate the abundance of flowering and to estimate the amount of pollen produced by two *Ligularia* species, which is available to pollinating insects as pollen forage. Pollen productivity of flowers and inflorescences of *L. clivorum* and *L. przewalskii* was compared. Pollen grain dimensions were determined, which can be useful for identification of pollen grains in microscopic analysis of bee products.

MATERIALS AND METHODS

Two perennial species of the *Ligularia* genus – *L. clivorum* Maxim. and *L. przewalskii* Maxim. (fig. 1A, B) were the object of the study. Under the conditions of south-eastern Poland (51°14'N and 22°34'E), observations of their flowering were performed in 2004–2008 and the rate of pollen production was estimated. For each taxon, the investigations were carried out for 3 growing seasons. In order to estimate the abundance of flowering, number of flower heads per stem and number of florets per flower head were determined, as well as plant density per unit area. Pollen production in flowers was examined by ether extraction and weight measurement following the method described by Warakomska [1972], using a modification that involved washing out the pollen initially with 70% ethanol, and subsequently with ether. In successive years of the study, 150 fully developed and mature anthers, just before dehiscence of their walls, were placed on each of six watch glasses. After the pollen was alternately dried and washed several times, it was weighed and the weight of pollen per 10 flowers, per inflorescence, and per unit area was determined.

In successive years of the study, pollen grains were mounted in glycerol-gelatine and then their dimensions were determined using an Eclipse 600 light microscope at a magnification of 40×15 . The length of polar (P) and equatorial (E) axes of the pollen grain as well as the P/E ratio were determined according to Erdtman [1954] classification.

The obtained results were statistically verified and the significance of differences between the means was compared by the Tukey test at a significance level of $\alpha = 0.05$.

RESULTS

Flowering of the studied taxa of the *Ligularia* genus took place during the summer time. The first inflorescences in *L. przewalskii* opened in the third decade of June and

they were in bloom almost until the end of July, from 29 to 38 days. The flowering period of *L. clivorum* started in the second or third decade of July and lasted 48 days in the first and third years of the study, while in the second year it was 67 days. The flower heads of *Ligularia* finished flowering latest in the first half of September (tab. 1).

Table 1. Period and flowering abundance of the examined species of *Ligularia*
Tabela 1. Termin i obfitość kwitnienia badanych gatunków *Ligularia*

Taxon Takson	Year Lata badań	Flowering Kwitnienie		Number of Liczba			
				flower heads per shoot koszyczków na pędzie		flowers per flower head (in total) kwiatów w koszyczku (ogółem)	
		period termin	number of days liczba dni	mean \pm SD średnia \pm SD	min.– max.	mean \pm SD średnia \pm SD	min.– max.
<i>L. clivorum</i>	2005	27.07–12.09	48	14.5 \pm 4.28	7–22	95.4 \pm 8.97	89.6–103.8
	2007	11.07–15.09	67	15.8 \pm 4.44	8–23	98.5 \pm 12.22	97.6–105.9
	2008	19.07–04.09	48	25.0 \pm 8.27	10–35	105.2 \pm 8.21	100.8–109.1
	mean średnia	-	54.3	18.4 ^B	-	100.6 ^A	-
<i>L. przewalskii</i>	2004	28.06–29.07	32	166.2 \pm 40.96	115–247	5.0 \pm 0.00	5.0
	2005	22.06–29.07	38	184.9 \pm 65.86	106–293	5.0 \pm 0.00	5.0
	2006	29.06–27.07	29	135.4 \pm 16.08	115–163	5.0 \pm 0.00	5.0
	mean średnia	-	33.0	162.2 ^A	-	5.0 ^B	-

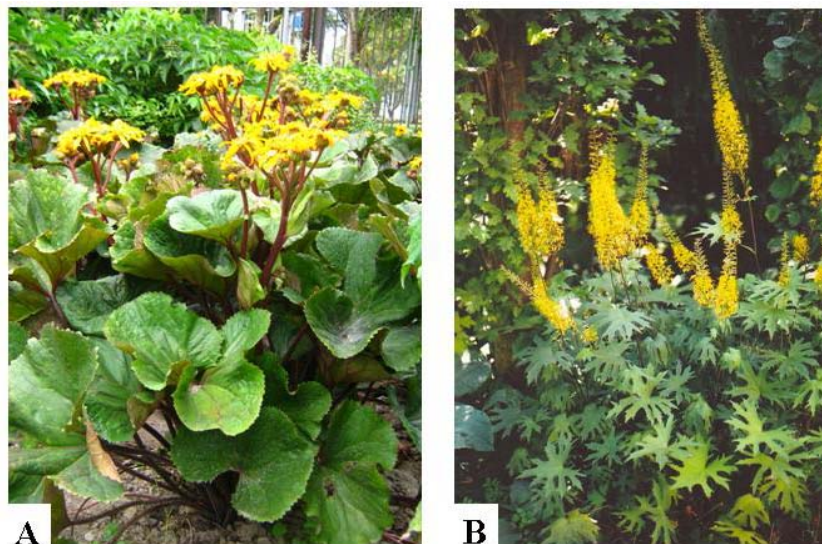
^{A, B} mean values within columns followed by different letters are significantly different at $\alpha = 0.05$

^{A, B} średnie wartości w kolumnach oznaczone różnymi literami różnią się istotnie dla $\alpha = 0,05$

Plants of both species produced basal inflorescence stems which reached a height from 82 to 105 cm at full bloom. They differ from each other in their morphological habit (phot. 1A, B). In *L. clivorum*, during the growing season an average of 18.4 flower heads are produced on one stem, which are borne in a loose apical panicle (tab. 1, phot. 1A). The capitula of *L. przewalskii*, numbering 162.2 per stem, form an impressive, spike-like, composite inflorescence (tab. 1, phot. 1B); its axis reached a length from 22 to 58 cm (on average 36.1 cm). Flowering duration for a flower head was strictly dependent on weather conditions; in *L. clivorum*, it lasted from 10 to 26 days, whereas in *L. przewalskii* from 4 to 7 days.

The diameter of *L. clivorum* flower heads reached an average value of 7.3 cm, whereas in *L. przewalskii* it was 3–4 times smaller. In both species, there are female ligulate florets on the edge of the receptacle, while its central part is occupied by bisexual tubular florets (phot. 2A, B).

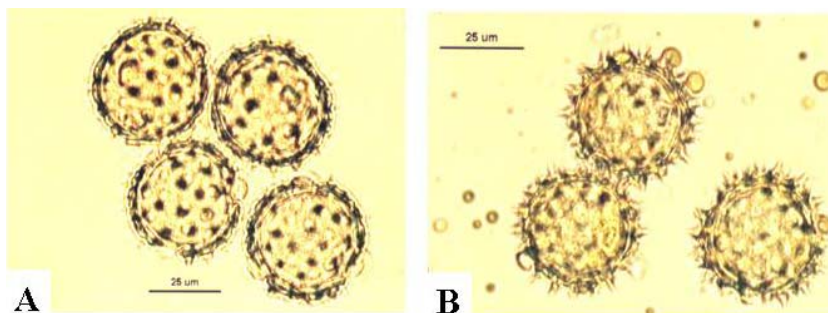
The flowering abundance of capitula significantly differed between the species under study. The total number of florets per *L. clivorum* inflorescence ranged 95.4–105.2, whereas in *L. przewalskii* this number was constant and amounted to 5.0. (tab. 1). In both taxa the proportions of particular types of florets in flower heads were similar in



Phot. 1. *Ligularia clivorum* Maxim. (A) and *Ligularia przewalskii* Maxim. (B) at full bloom
 Fot. 1. *Ligularia clivorum* Maxim. (A) i *Ligularia przewalskii* Maxim. (B) w pełni kwitnienia



Phot. 2. Flower heads with ligulate and tubular flowers of *L. clivorum* (A) and *L. przewalskii* (B)
 Fot. 2. Koszyczki z kwiatami języczkowatymi i rurkowatymi *L. clivorum* (A) i *L. przewalskii* (B)



Phot. 3. Microscopic view of pollen grains – *L. clivorum* (A) and *L. przewalskii* (B)
 Fot. 3. Obraz mikroskopowy ziaren pyłku – *L. clivorum* (A) i *L. przewalskii* (B)

the years of the study. In *L. clivorum*, tubular florets predominated by far, with their percentage standing at 86.2–87.1% (on average 86.7%), while in *L. przewalskii* their proportion was constant and it was 60.0% (fig. 1).

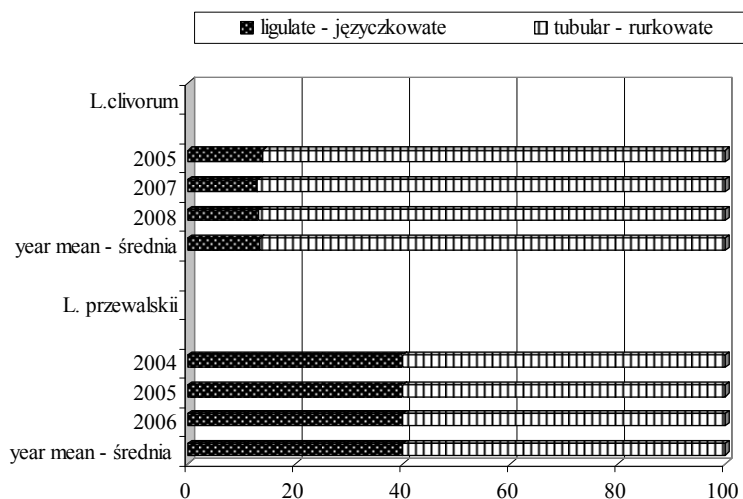


Fig. 1. Contribution of various flowers types in flower head (%)

Ryc. 1. Udział różnych typów kwiatów w koszyczku (%)

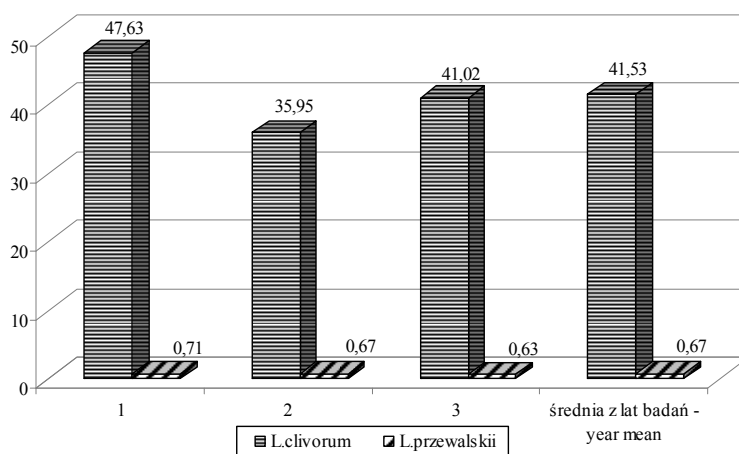


Fig. 2. Pollen weight per flower head of *Ligularia* (mg) in the years of study (1–3)

Ryc. 2. Masa pyłku z jednego koszyczka *Ligularia* (mg) w latach badań (1–3)

Similarly to other representatives of the Asteraceae family, the development of florets in *Ligularia* flower heads proceeds from the edge of the receptacle towards its central part. These flowers are typically entomophilous and are characterized by the earlier maturation of anthers than of the pistil – which is called protandry.

Pollen grains are produced only in tubular florets, which are characterized by the presence of fertile stamens. The dehiscence of the anther walls and pollen release begin already at the loose bud stage, just before the corolla lobes open. The pollen is accessible to insects only after it is pushed out of the staminal tube by the stigmas of the inferior pistil, which are protruding from the tube.

The weight of pollen produced by 10 flowers differed significantly in the studied species and averaged 4.26 mg for *L. clivorum* and almost twice less, 2.23 mg, for *L. przewalskii* (fab. 2). Pollen yield per flower head in *L. clivorum* was many times higher, which was attributable to more abundant pollen production in the stamens and a much higher number of tubular florets in the flower heads of this species (fab. 1, 2; fig. 2). The estimated average pollen production per stem and per unit area (1 m²) was, respectively: for *L. clivorum* 727.88 mg and 6.92 g, for *L. przewalskii* 109.06 mg and 1.53 g (fab. 2).

Table 2. Pollen productivity of *Ligularia*
Tabela 2. Wydajność pyłkowa *Ligularia*

Taxon Takson	Years Lata badań	Mean mass of pollen/ Średnia masa pyłku/		
		10 flowers (mg) ± SD 10 kwiatów (mg) ± SD	1 shoot – 1 pędu (mg)	1 m ² (g)
<i>L. clivorum</i>	2005	4.37 ± 1.70	747.52	7.10
	2007	4.55 ± 0.85	505.92	4.81
	2008	3.85 ± 0.75	930.21	8.84
	mean średnia	4.26 ^A	727.88	6.92
<i>L. przewalskii</i>	2004	2.37 ± 0.11	118.00	1.65
	2005	2.23 ± 0.20	123.88	1.73
	2006	2.09 ± 0.17	85.30	1.19
	mean średnia	2.23 ^B	109.06	1.53

^{A, B} mean values within columns followed by different letters are significantly different at $\alpha = 0.05$

^{A, B} średnie wartości w kolumnach oznaczone różnymi literami różnią się istotnie dla $\alpha = 0,05$

Pollen grains of *Ligularia*, roundish in outline, tricolporate, are characterized by echinate exine sculpturing (phot. 3A, B). Their P/E ratio reached mean values of 0.98 and 1.04. The average length of the polar and equatorial axes of the pollen grain and the length of spines (in μm) were as follows: 38.26, 36.89, and 3.63 in *L. clivorum*; 34.49, 35.33, and 6.45 in *L. przewalskii* (tab. 3).

Table 3. Dimensions of *Ligularia* pollen grains
Tabela 3. Wymiary ziaren pyłku *Ligularia*

Taxon Takson	Years Lata badań	Length of axis (μm) – Długość osi				Mean length of spikes (μm) \pm SD Średnia długość kolców (μm) \pm SD		Shape index P/E
		polar (P) – biegunowej (P) mean \pm SD średnia \pm SD	min. – max.	equatorial (E) – równikowej (E) mean \pm SD średnia \pm SD	min. – max.	(μm) \pm SD	(μm) \pm SD	
<i>L. chivorum</i>	2005	36.62 \pm 2.26	33.85–44.09	35.15 \pm 2.08	31.41–39.18	3.59 \pm 0.43	1.04	
	2007	39.38 \pm 1.45	36.68–41.59	38.39 \pm 1.34	36.07–41.11	4.07 \pm 0.42	1.02	
	2008	38.77 \pm 1.90	33.98–42.87	36.85 \pm 2.05	28.91–41.54	3.22 \pm 0.61	1.05	
	mean średnia	38.25 ^A	-	36.80 ^A	-	3.63 ^B	1.04	
<i>L. przewalskii</i>	2004	35.89 \pm 2.24	31.25–40.25	37.09 \pm 2.39	32.06–42.06	6.61 \pm 0.61	0.97	
	2005	33.13 \pm 1.39	31.09–34.51	34.05 \pm 1.90	31.17–39.54	6.58 \pm 0.76	0.97	
	2006	34.44 \pm 2.00	30.62–39.41	34.85 \pm 1.65	32.12–39.41	6.16 \pm 0.62	0.99	
	mean średnia	34.49 ^B	-	35.33 ^B	-	6.45 ^A	0.98	

^{A, B} mean values within columns followed by different letters are significantly different at $\alpha = 0.05$
^{A, B} średnie wartości w kolumnach oznaczone różnymi literami różnią się istotnie dla $\alpha = 0.05$

DISCUSSION

Under the conditions of south-eastern Poland, flowering of both *Ligularia* species lasts, depending on species and weather conditions, from 4 to more than 9 weeks. The inflorescences of *L. przewalskii* started to flower 2–3 weeks earlier than it was in the case of *L. clivorum*. The time of flowering of *L. clivorum* is in agreement with the one given by Radziul [2002] for Polish conditions, whereas according to Rutkowski [1998] and Marcinkowski [2002], this taxon blooms as late as August, and the end of its flowering is observed in September, which was confirmed by the present study. Lipiński [2010] describes *L. clivorum* as a bee forage species of the second half of summer.

At the flowering stage, the plants of the studied taxa grew up to a height of about 1 m (82–105 cm). These values are lower than those cited by Radziul [2002] and Marcinkowski [2002], according to whom inflorescence stems of the taxa under study can grow up even to a height of 150 cm. These authors also report larger dimensions of the axis length of the spike-like composite inflorescence of *L. przewalskii*; in our study, this length reached 22–58 cm.

The capitula of both *Ligularia* species differ from each other in their morphological structure. In *L. przewalskii*, flower heads are several times smaller and contain only 5 florets, whereas in *L. clivorum* the number of florets per head is 20–25 times higher. Among these two types of florets distinguished in the inflorescences of the studied taxa, bisexual tubular florets were predominant and they were characterized by the presence of fertile stamens. In *L. clivorum*, their proportion in a flower head was higher (86.7%) compared to *L. przewalskii* (60.0%) in the case of which the number of florets per inflorescence was a constant value. Wróblewska (1995) observed the presence of the same types of florets, as in *Ligularia*, in the flower heads of *Tagetes erecta* and *Tagetes patula* which contained 59.4% and 58.9%, respectively, of pollen-producing tubular florets.

Pollen forage is provided exclusively by tubular florets whose anthers are fused together in a tube; pollen grains are accumulated inside this tube after they are matured and released from the anthers. Insects can forage on the pollen of *Ligularia* only after it has been pushed outside the staminal tube by the stigmas of the pistil. A similar mechanism of pollen maturation and release has been observed by Wróblewska [1997] in *Silphium perfoliatum* and by Wróblewska and Magacz [2006] in *Zinnia elegans*.

Compared to the pollen grains of *L. przewalskii*, the pollen grains of *L. clivorum* are characterized by slightly larger dimensions and much shorter spines. In both species, they are included in the group of medium-sized grains, since their dimensions are in the range of 25–50 μm [Erdtman 1954]. In their morphological descriptions of representatives of the Asteraceae family, Chiguryayeva and Tereshkova [1998] as well as Punt and Hoen [2009] include the pollen grains of *L. clivorum* in the type of *Ligularia sibirica*, which comprises pollen grains with the dimension of the axis of more than 35 μm .

The results on pollen productivity of *Ligularia* flowers have no equivalents in the literature. The average pollen weight per 10 *L. clivorum* flowers was almost twice higher than that in *L. przewalskii* in the study period. The value of pollen weight per 10 *L. clivorum* flowers (4.26 mg) is close to the one estimated for *Silphium perfoliatum*

[Wróblewska 1997], *Centaurea cyanus* [Denisow 2006] and it is higher than in *Calendula officinalis*, *Cosmos bipinnatus*, and *Tagetes patula* [Wróblewska 1995]. An average of 2.23 mg of pollen was obtained from 10 *L. przewalskii* flowers, which is a higher value than that given by Warakomska and Kolasa [2003] for *Tussilago farfara*, Wróblewska and Magacz [2006] for *Zinnia elegans* as well as by Strzałkowska [2006] for several *Solidago* species.

Pollen weight, calculated per flower head, recorded for *L. clivorum* exceeded many times the values obtained for *L. przewalskii*, which resulted from a significantly higher number of tubular (pollen-producing) florets in the inflorescences of *L. clivorum*. According to Percival [1955], from 2.1 mg (*Senecio jacobaea*) to 239.0 mg (*Helianthus annuus*) of pollen can be obtained from a single flower head of various species of the Asteraceae family. The data published by Denisow [2011] show that representatives of this family in ruderal communities of the city of Lublin can provide from 0.06 mg (*Artemisia vulgaris*) to 49.54 mg (*Onopordum acanthium*) of pollen per inflorescence. The weight of pollen produced is also affected by the dimensions of pollen-producing flowers in the inflorescence and weather conditions which determine the abundance of flowering in flower heads.

CONCLUSIONS

1. The studied species of the *Ligularia* genus flower abundantly under the conditions of south-eastern Poland during the summer period: *L. clivorum* blooms for 48–67 days, *L. przewalskii* for 29–38 days.

2. Two types of flowers in the flower heads of both species are found – female ligulate florets and bisexual tubular florets. Their proportions in the inflorescences reached, respectively: 13.3% and 86.7% in *L. clivorum*, 40.0% and 60% in *L. przewalskii*.

3. The weight of pollen produced by 10 flowers was on average 4.26 mg for *L. clivorum* and 2.23 mg for *L. przewalskii*. Respectively, 6.92 g and 1.53 g of flower pollen, which is a valuable protein food for pollinating insects, can be obtained from a 1 m² area of a dense stand of both species.

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OBFITOŚĆ KWITNIENIA I WYDAJNOŚĆ PYŁKOWA *Ligularia clivorum* Maxim. I *Ligularia przewalskii* Maxim.

Streszczenie. Rodzaj *Ligularia* skupia gatunki uprawiane w ogrodach przydomowych i na rabatach jako atrakcyjne rośliny ozdobne. W okresie kwitnienia ich koszyczki kwia-

towe mogą dostarczać pokarmu pyłkowego dla zapylaczy. W latach 2004–2008 badano intensywność kwitnienia oraz pylenia dwu gatunków *Ligularia* jako źródeł pożytku pyłkowego dla owadów. W warunkach Lublina początek kwitnienia *L. clivorum* przypada w drugiej połowie lipca, *L. przewalskii* już w ostatnim tygodniu czerwca. W zależności od gatunku i czynników pogody, okres kwitnienia *L. clivorum* trwał 48–67 dni, *L. przewalskii* od 29 do 38 dni. W kwiatostanach typu koszyczka znajdują się u obu taksonów kwiaty języczkowe żeńskie i rurkowane obupłciowe. Masa pyłku wytworzona przez 10 kwiatów *L. clivorum* była prawie dwukrotnie wyższa niż u *L. przewalskii*. Wydajność pyłkowa z jednostki powierzchni osiągnęła dla tych taksonów odpowiednio 6,92 g·m⁻² i 1,53 g·m⁻².

Słowa kluczowe: Asteraceae, kwitnienie, wydajność pyłkowa

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