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## Honey bee foraging in spring oilseed rape crops under high ambient temperature conditions

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### Abstract

Honey bee foraging activity on the flowers of two spring rapeseed varieties ‘SW Savann’ and ‘Ural’ was evaluated. High air temperature throughout the study period allowed us to investigate the interaction between plants and their pollinators under weather conditions unusual for Lithuania.

Analysis of flowering intensity and honey bee density in the two rape varieties showed that ‘Ural’ produced on average 4.6% more flowers than ‘SW Savann’, however, honey bee density in ‘Ural’ plots was about 4% lower than that in ‘SW Savann’ plots. A decrease in flowering intensity was followed by a decrease in honey bee density in both rape varieties. A strong increase in ambient temperature had a negative impact on the foraging of honey bees on flowering plants. The lowest honey bee density in the investigated rape plots was recorded in the afternoon, when air temperature reached +43°C.

High ambient temperature affected oilseed rape flowering and pollinator density on flowers and this could have had a negative effect on seed yield of oilseed rape.

Key words: oilseed rape, *Brassica napus*, honey bee, *Apis mellifera*, pollination, ambient temperature.

### Introduction

Oilseed rape (*Brassica napus* L.) is widely grown for food and animal feed. This valuable oilseed crop has a growing demand in the world market because of its use in biofuel production. In Lithuania, the cultivation of rape is also gradually increasing.

Bright yellow rape flowers containing abundant pollen and nectar are attractive to many insect pollinators that, in turn, contribute to a better seed set and the yield of rape (Burgett et al., 1993; Langridge, Goodman, 1996; Steffan-Dewenter, 2003; Koltowski, 2005). Honey bees are the main pollinators of oilseed rape, accounting for from 46% to 95% of all insect pollinators of this crop (Mesquida et al., 1988; Blight et al., 1997; Koltowski, 2001; Pierre et al., 2003). Other rape pollinators, such as solitary bees can account for about 4% or some-

times 9% of all insect pollinators (Koltowski, 2001). Bumble bees being important pollinators of many agricultural crops, however, make up only 2% of all insect pollinators in rape crops (Cresswell, 1999; Koltowski, 2001). The attractiveness of plants to pollinators depends on a variety of factors. Climate changes due to global warming are assumed to have impact on the already established mutualistic relationships between flowering plants and insect pollinators (Blažytė-Čereškienė, 2007, review).

It should be noted that in Lithuania more and more often we witness climate changes that are related to global warming, i.e. warmer winter temperatures and longer periods of hot weather in summer. More frequent losses of winter rape crops both in Lithuania and neighbouring countries (Koltowski, 2001) encouraged farmers to focus more on spring varieties.

The aim of this study was to determine honey bee foraging activity and the peculiarities of its dynamics in two spring rapeseed varieties ‘Ural’ and ‘SW Savann’. High air temperature during the study period allowed us to investigate the interaction between plants and their pollinators under weather conditions unusual for Lithuania.

## Materials and methods

**Rape varieties.** Investigations were carried out in the Agricultural Company “Atžalynas” (Makančiškė village, Lazdijai distr., Lithuania) on 8–12 July, 2006. Two rape varieties (‘Ural’ and ‘SW Savann’) were used. The total area of rape crop was 60 ha. ‘SW Savann’ made up 1/4, whereas ‘Ural’ 3/4 of the total crop area.

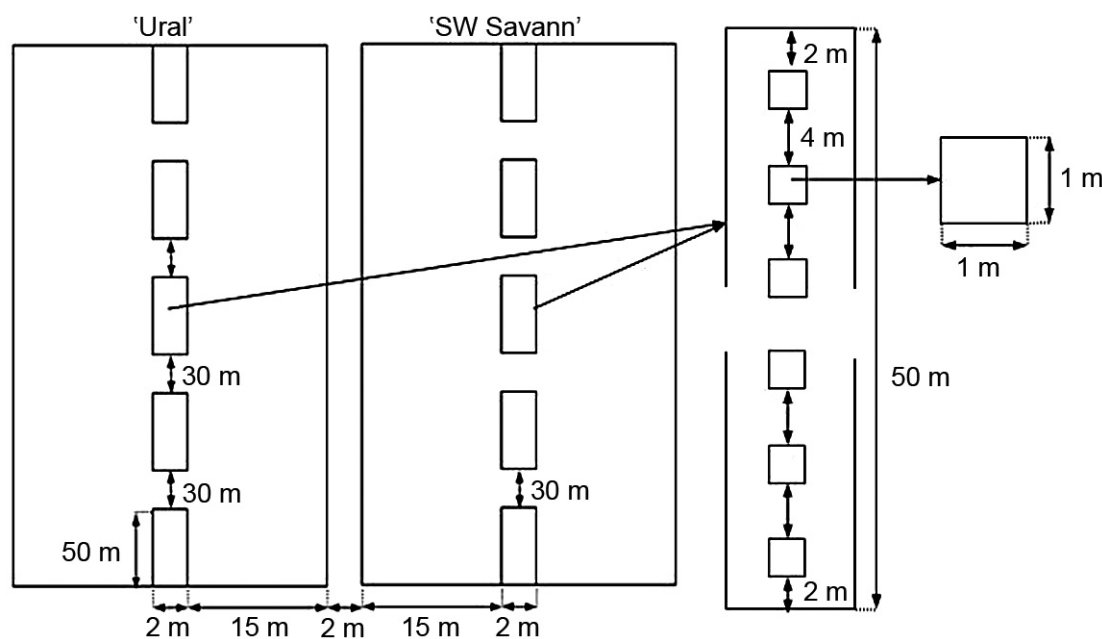
‘Ural’ is an early highly productive rapeseed variety producing medium-sized oilseeds, with an average 1,000-seed weight of 3.7 g. Its medium-height seedlings are resistant to lodging and seeds are firmly attached to siliquae. It is also disease-resistant and more tolerant to poor soils. The mid-early ‘SW Savann’ variety is also highly productive and produces large oilseeds, with an average 1,000-seed weight of 3.95 g. The plants of this variety are tall, resistant to lodging; seeds are firmly attached to siliquae (Lithuanian State..., 2009). Both varieties,

‘Ural’ since 2001 and ‘SW Savann’ since 2005, have been included in the list of plants suitable to be grown in Lithuania.

**Study plots and environment.** The scheme of study plots was established as presented in Fig. 1. The crop fields of both varieties were separated by a 2 m wide unseeded path. Ten study plots (2 x 50 m) were established along each crop field at a distance of 15 m from this path. Five study plots were set up for each variety. The observation area made up 1 ha of the total crop field.

Ambient temperature was measured regularly three times a day at the level of flowers using a glass thermometer.

**Evaluation of plant and flower density.** To evaluate the flower density of rape, the middle study plot in each investigated crop area was selected and divided into 10 smaller plots (1 x 1 m) as shown in Fig. 1. On the first day of investigation, the number of plants for each variety was calculated. Assuming that the number of plants remained stable throughout the investigation period, the mean density of plants for each variety was calculated. In each 1 m<sup>2</sup> plot, separated by a distance of 4 m, the flowers on 10 randomly selected plants were counted. The counting was performed at 1–4 p.m. three times during the investigation period: on 8, 10 and 12 of July.



Note. Insects were counted in large plots 2 x 50 m (left). Ten small plots 1 x 1 m (right) were set up in each middle-sized plot for flower counting.

**Figure 1.** Experimental design of rapeseed plots

To evaluate rape flower density, the number of flowers per 1 m<sup>2</sup> of the crop area was counted according to the following formula:

$$F = \sum_{i=1}^n P_i / 10 \times N, \text{ where}$$

$F$  – number of flowers/1 m<sup>2</sup> of the crop area,  $n$  – number of plants selected randomly for flower count ( $n = 10$ ),  $P$  – number of flowers per plant,  $N$  – number of plants/1 m<sup>2</sup> of the crop area.

The flowering of rape varieties was compared based on the number of flowers and the percentage of flowers per 1 m<sup>2</sup> of the crop area. The number of flowers in all plots of both varieties during the first day of observation was counted as 100%.

The dynamics of flowering during the investigation period were evaluated based on the percentage of flowers per 1 m<sup>2</sup> of the crop area.

*Evaluation of insect density.* The density of honey bees was determined by counting foraging insects observed by walking along the predefined study plots. Observations were made in ten plots. Area of each plot was 100 m<sup>2</sup> (Fig. 1, left). In the plots, we counted only the honey bees landed on flowers. An insect in the same study plot that landed on the flower more than once was considered as new individual. The number of honey bees for each variety was counted 15 times: three times a day over the period of 5 days at the same time (in the morning from 9 a.m. to 10 a.m., in the noon from 12 a.m. to 1 p.m. and in the afternoon from 4 p.m. to 5 p.m.). The observation was made by walking along the one 100 m<sup>2</sup> study plot lasted for 5 min. Insect observation in all ten plots of both varieties lasted for about 1 hour every day.

Honey bee density (the number of honey bees per 100 m<sup>2</sup> of the crop area) and the dynamics of insect density during the day and during the investigation period were evaluated.

*Statistical data analysis.* Nonparametric tests were used (Čekanavičius, Murauskas, 2002) to evaluate and compare the results obtained. The Friedman test was used to evaluate flowering dynamics and honey bee density during the day and throughout the investigation period because the observation was made in the same plots, but at a different time. The Wilcoxon matched pairs test was used to assess differences in flowering and honey bee density between two observations in the plots

of the same variety. Differences in flower and honey bee density between the two varieties were evaluated using the nonparametric Mann-Whitney U-test, whereas the correlation between the daily dynamics of insect and flower densities and changes in temperature was determined using the Spearman rank test.

## Results

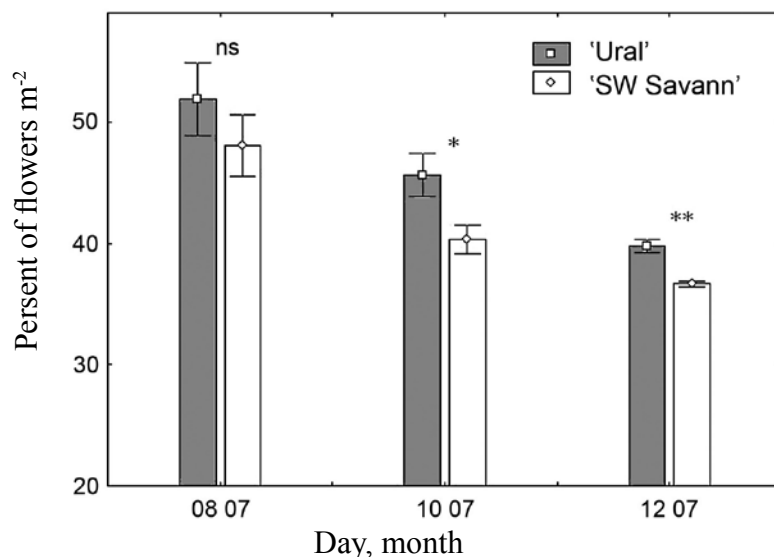
*Climatic conditions.* During the 5-day investigation period, the weather was windless (wind speed was up to 3 m/s) and sunny, except for cloudy afternoons of the second and third day of investigation. High temperature persisted over all days and the observations were made under extreme climate conditions, unusual for Lithuania. During the morning observation, air temperature fluctuated from +27°C to +30°C ( $28.2 \pm 0.17$ ), during the noon from +34°C to +42°C ( $37.2 \pm 0.42$ ), whereas during the afternoon varied from +39°C to +45°C ( $43.0 \pm 0.30$ ), reaching the peak. The highest mean temperature was recorded during the afternoon observation. The lack of precipitation and high temperature resulted in soil cracking and wilting of plants that recovered only the next morning.

*Rape flowering and its dynamics.* No differences in plant density were found between the two varieties ( $p > 0.05$ , Mann-Whitney U-test). The mean density was  $59.1 \pm 0.90$  plants/m<sup>2</sup> for ‘SW Savann’ and  $55.9 \pm 1.12$  plants/m<sup>2</sup> for ‘Ural’. Significant differences were recorded in average number of flowers produced per plant of both varieties. One plant of ‘SW Savann’ produced on average  $18.1 \pm 0.55$  flowers, whereas that of ‘Ural’  $19.9 \pm 0.64$  flowers ( $p = 0.009$ , Mann-Whitney U-test). Comparison of flower density in the crop area revealed no differences between the two varieties ( $p > 0.05$ , Mann-Whitney U-test). However, ‘Ural’ produced on average 4.6% more flowers than ‘SW Savann’ ( $p < 0.001$ , Mann-Whitney U-test).

The density of flowers during the short observation period decreased significantly in both varieties (‘Ural’:  $\chi^2 = 9.4$ ,  $N = 10$ ,  $df = 2$ ,  $p < 0.009$ ; ‘SW Savann’:  $\chi^2 = 12.8$ ,  $N = 10$ ,  $df = 2$ ,  $p < 0.002$ ; Friedman test) (Fig. 2). On the first observation day, the flowering in both varieties was highest and very similar. ‘SW Savann’ produced on average  $1244.6 \pm 83.62$  flowers and ‘Ural’  $1266.0 \pm 97.00$  flowers/m<sup>2</sup> of the area. On the second observation day, the flowering in both rape varieties was lower. Moreover, ‘Ural’

produced significantly (5.3%) more flowers than 'SW Savann' ( $p = 0.017$ , Mann-Whitney U-test). The lowest flowering was observed on the last day, accounting on average for  $941.1 \pm 24.49$  flowers/m<sup>2</sup>

in 'SW Savann' and  $965.4 \pm 31.69$  flowers/m<sup>2</sup> in 'Ural'. On this day, differences in flowering made up only 3.2%, but they were statistically significant ( $p < 0.001$ , Mann-Whitney U-test).



**Figure 2.** Dynamics of flowering over the 5-day period (differences not significant (ns):  $p > 0.05$ ; differences significant: \* –  $p < 0.05$ , \*\* –  $p < 0.001$ ; Mann-Whitney U-test)

**Dynamics of honey bee activity in two rape varieties.** The foraging activity of honey bees evaluated as percentage of insects in the crop field in the two rape varieties was different. 'SW Savann' flowers were visited by honey bees 4% more often than 'Ural' flowers ( $p < 0.001$ , Mann-Whitney U-test).

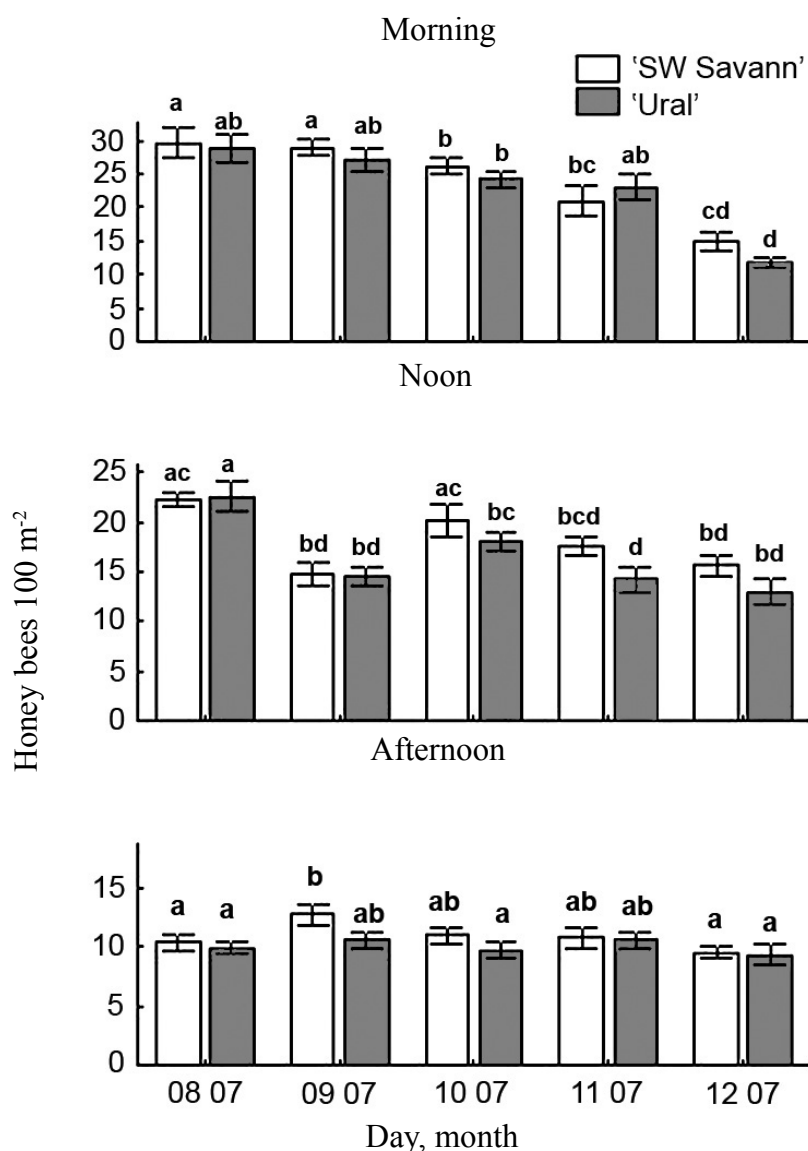
The foraging activity of honey bees in the plots of both varieties depended on the time (morning, noon, afternoon) and the day of observation (Fig. 3).

During the 5-day investigation period, honey bee density over the day decreased significantly in the crop fields of both varieties ( $p < 0.05$ , Friedman test), except during the last observation day in the 'Ural' crop, when no statistically significant differences in honey bee density were found between morning, noon and afternoon observations ( $\chi^2 = 2.8$ ,  $N = 5$ ,  $df = 2$ ,  $p > 0.05$ ; Friedman test). The largest number of honey bees in both varieties was found during the morning observation (on average  $23.6 \pm 0.95$  insects/100 m<sup>2</sup>) and the smallest number in the afternoon ( $10.5 \pm 0.25$  insects/100 m<sup>2</sup>).

Though a decline in honey bee density in the afternoon was observed every day over the whole 5-day period, a pronounced decrease was re-

corded on the first day (on average from  $29.2 \pm 1.44$  to  $10.2 \pm 0.42$  insects/100 m<sup>2</sup>). The results revealed a gradual daily decrease in honey bee density in both rape varieties (Fig. 3).

In the 'SW Savann' plots, honey bee density on the first day during the morning observation accounted for  $29.6 \pm 2.29$  insects/100 m<sup>2</sup>, whereas on the last day of investigation for  $15.0 \pm 1.30$  insects/100 m<sup>2</sup>. This decline was statistically significant ( $\chi^2 = 15.6$ ,  $N = 5$ ,  $df = 2$ ,  $p < 0.0036$ ; Friedman test). The same trend of a statistically significant decrease in honey bee density ( $\chi^2 = 13.9$ ,  $N = 5$ ,  $df = 2$ ,  $p > 0.008$ ; Friedman test) was also found during the noon observation. The highest density ( $22.2 \pm 0.66$  insects/100 m<sup>2</sup>) was recorded on the first day, whereas on the last day of investigation a significant decrease of  $15.6 \pm 1.03$  insects/100 m<sup>2</sup> was observed. The lowest density variation was during the afternoon observation and varied between  $12.8 \pm 0.86$  insects/100 m<sup>2</sup> and  $9.6 \pm 0.51$  insects/100 m<sup>2</sup>. Statistical analysis revealed no significant differences in honey bee density in the afternoon during the investigation period ( $\chi^2 = 8.7$ ,  $N = 5$ ,  $df = 2$ ,  $p > 0.05$ ; Friedman test).



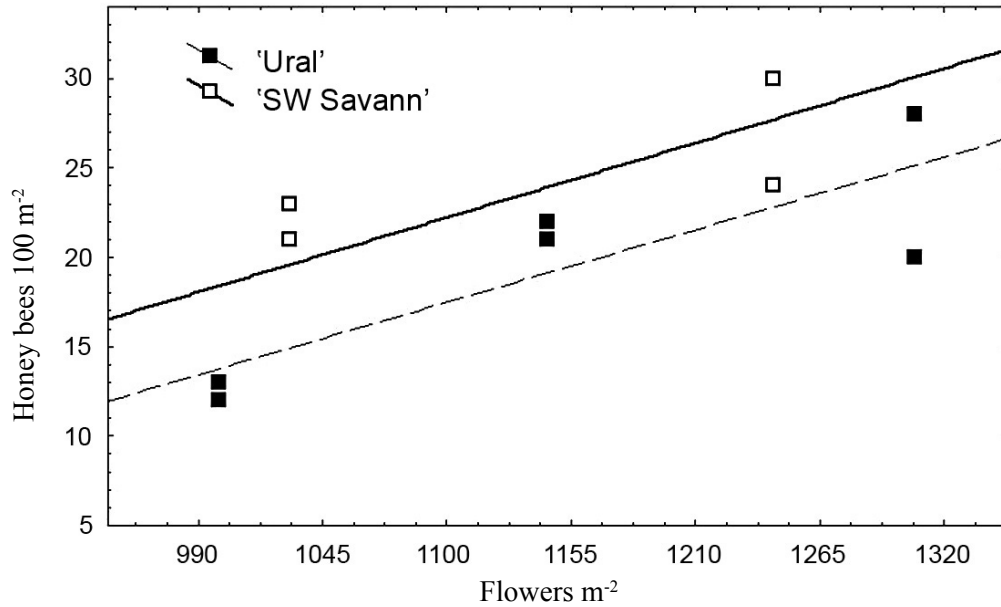
Note. Different letters (a, b, c, d) denote significant differences ( $p < 0.05$ , Mann-Whitney U-test and Wilcoxon matched pairs test).

**Figure 3.** Dynamics of honey bee density (insects/100 m<sup>2</sup>) in the 'SW Savann' and 'Ural' rape varieties during the investigation period

The same dynamics of honey bee density were found for the 'Ural' variety. Comparison of honey bee density during the morning observation over the 5-day period revealed significant differences ( $\chi^2 = 13.8$ ,  $N = 5$ ,  $df = 2$ ,  $p < 0.008$ ; Friedman test). Honey bee density varied from  $28.8 \pm 1.98$  insects/100 m<sup>2</sup> to  $11.8 \pm 0.58$  insects/100 m<sup>2</sup>. Statistically significant differences in honey bee density were also found during the noon observation ( $\chi^2 = 13.6$ ,  $N = 5$ ,  $df = 2$ ,  $p < 0.009$ ; Friedman test). On the first day of investigation, honey bee density reached  $22.6 \pm 1.54$  insects/100 m<sup>2</sup>, whereas on the last day  $13.0 \pm 1.34$  insects/100 m<sup>2</sup>. No differences were observed in density during the afternoon ob-

servation (4 p.m.) over the 5-day period ( $\chi^2 = 1.3$ ,  $N = 5$ ,  $df = 2$ ,  $p > 0.05$ ; Friedman test).

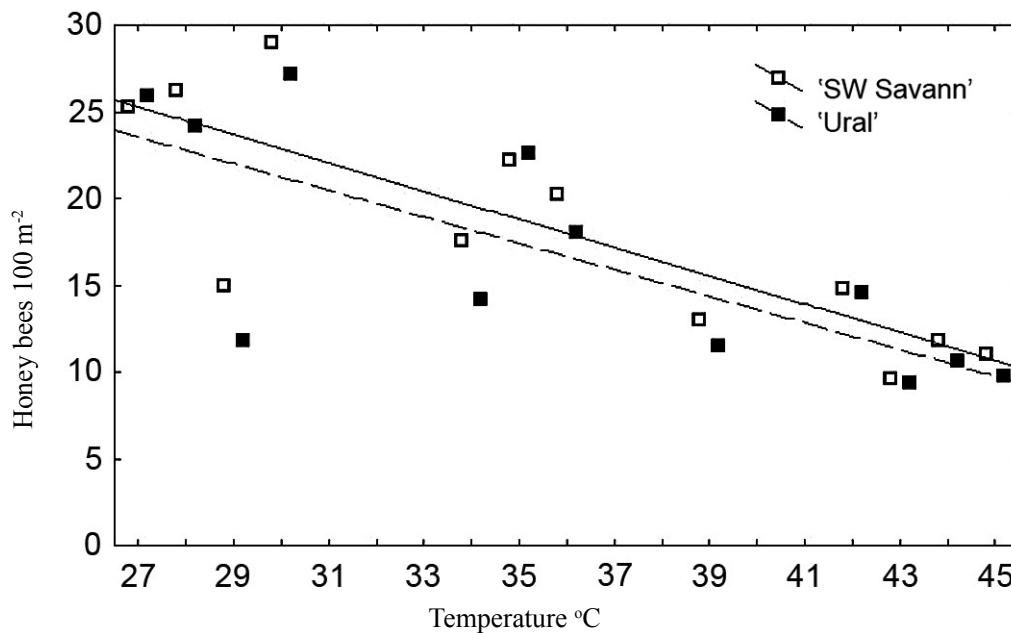
**Relation between honey bee density and flower density.** Variation in honey bee density over the 5-day period may have been due to a significantly decreased density of flowers. Spearman correlation allowed us to evaluate the relationship between insect and flower densities. Honey bee density in the investigated rape varieties were positively correlated with flower density ('Ural' –  $R = 0.72$ , 'SW Savann' –  $R = 0.95$ ; Spearman correlation). Thus, an increase in flowering intensity was followed by an increase in honey bee density in both rape varieties (Fig. 4).



**Figure 4.** Correlation between honey bee density and flower density in the 'SW Savann' and 'Ural' rape varieties ('Ural' –  $R = 0.72$ , 'SW Savann' –  $R = 0.95$ ; Spearman correlation)

**Relation between honey bee density and ambient temperature.** Comparison of trends in the dynamics of temperature and honey bee density indicated that honey bee density was correlated with ambient temperature in both rape varieties

investigated ('Ural' –  $R = 0.75$ , 'SW Savann' –  $R = 0.76$ ; Spearman correlation). Consequently, an increase in temperature was followed by a decrease in honey bee density in both rape varieties (Fig. 5).



**Figure 5.** Correlation between mean honey bee density and ambient temperature ('SW Savann' –  $R = -0.76$ , 'Ural' –  $R = -0.75$ ; Spearman correlation)

## Discussion

The results obtained revealed some slight differences in the flowering intensity of the two oilseed rape varieties and honey bee foraging activity on the flowers of these varieties (Fig. 2). In total, 'Ural' produced 4.6% more flowers compared to 'SW Savann', but 'Ural' flowers were less visited by honey bees than 'SW Savann' flowers.

Koltowski (2001; 2005) investigating winter rape found some differences in honey bee density on different rape varieties, too. The activity of nectar foraging honey bees depends on many factors, among which nectar availability (quantity, quality, and accessibility) is important. Honey bees are able to discriminate among rape genetic lines that give more or less reward (Gupta et al., 1984; Picard-Nizou et al., 1995) and they adapt the number of their visits to the nectar secretion rate (Williams, 1997). There are reports that the amount of sugars secreted in the nectar of different oilseed rape varieties varied (Bobrzecka, Bobrzecki, 1973; Koltowski, 2001). In our study, the rape varieties ('SW Savann' and 'Ural') were not similar in attractiveness or in accessibility of nectar to honey bees, therefore insects visited 'SW Savann' variety more often than 'Ural'. It is likely that 'SW Savann' flowers produced a better quality or quantity of nectar compared to 'Ural' flowers.

Comparison of the dynamics of honey bee density in the investigated spring rape varieties revealed the same diminishing trend for both varieties both over the day and over the whole 5-day period (Fig. 3). The highest density of honey bees in the plots of both rape varieties was recorded in the morning, whereas the lowest in the afternoon. Many authors note the diminished activity of honey bees in the afternoon (Winston, 1987; Danka et al., 2006). The reasons for this phenomenon are not clear, though, at least for the summer period, a certain relationship may exist between honey bee activity and solar radiation (Butler, Finney, 1942; Abrol, 2006), relative humidity and temperature (Gary, 1992).

Under environmental conditions which are common in the summer season in Lithuania, honey bee foraging activity until afternoon is usually high (Butler, Finney, 1942; Danka et al., 2006). The diminished density of honey bees during the noon (12 a.m.) observation in our study might be related to increase in ambient temperature to +36–42°C. Ambient temperature is often considered to be an

important factor determining the relationship between the flowering plant and honey bee behaviour (Danka et al., 2006; Kreitlow, Tarpy, 2006; Afik, Shafir, 2007). During the present research period, temperature in the morning was the lowest (+28°C), whereas in the afternoon rose up to +43°C. Comparison of trends in the dynamics of temperature and honey bee density indicated that an increase in temperature was followed by a decrease in honey bee density (Fig. 5). Our data are in accordance with studies reporting that extreme temperature affects the pollen collection activity of honey bees. The optimal temperature for pollen collection in Cape honey bees was between +14°C to +26°C, whereas their activity decreased when temperature rose above +26°C (Adamssu, 2003). A decline in flight activity was observed for temperatures above +30°C, honeybees remained at their hive entrance presumably ventilating the hives to reduce hive temperatures (Adamssu, 2003; Danka et al., 2006). Furthermore, high temperature influences floral scent production and emission (Sagae et al., 2008), and nectar secretion in the plant (Petanidou, Smets, 1996). Based on these data and our results, we conclude that an increase in temperature had a negative impact on the foraging of honey bees on flowering plants, both due to direct effect on the honey bees and indirect effect through the plant flowering, scent and nectar production decline.

Along with unusually high ambient temperature for the middle latitude, the intensity of flowering is another important factor that might have affected honey bee activity in the investigated rape plots. The unusually high temperature led to a rapid (over 5 days) decrease in the number of flowers (Fig. 2). A decrease in the number of flowers in both varieties was followed by a decrease in the density of honey bees (Fig. 4). Especially significant changes in the number of honey bees were found during the morning observations, when at the end of the investigation period honey bee density in the rape plots decreased twice.

Consequently, unusually high ambient temperature might have had both direct and indirect effect on the activity of honey bees and thus determined their density in the rape plots. Moreover, a shorter flowering period and a low density of pollinators due to extremely high ambient temperature could have had a negative effect on the seed yield of oilseed rape.

## Conclusions

Analysis of honey bee activity on the flowers of 'SW Savanna' and 'Ural' rape varieties showed that:

1. 'Ural' produced on average 4.6% more flowers than 'SW Savanna', however, the activity of honey bees in 'Ural' plots was about 4% lower than in 'SW Savanna' plots.

2. A decrease in flowering intensity was followed by a decrease in honey bee density in both rape varieties.

3. An extreme increase in ambient temperature had a negative impact on the foraging of honey bees on flowering plants. The lowest honey bee density was recorded in the afternoon, when air temperature reached +43°C.

4. A shorter flowering of oilseed rape and a low density of pollinators on flowers due to extremely high ambient temperature could have had a negative effect on the seed yield of oilseed rape.

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## Medunešių bičių gausumas vasarinių rapsų pasėliuose aukštos aplinkos temperatūros sąlygomis

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### Santrauka

Rapsas (*Brassica napus*) – plačiai ir nuo seno auginama vertinga aliejinė kultūra, kuri pastaraisiais metais tapo svarbi ekologiniu atžvilgiu, kai buvo pradėta biokuro gamyba iš rapsų sėklų, lėmusi jų poreikio pasaulio rinkoje spartų augimą. Rapsų pasėliuose didelį sėklų derlių gali užtikrinti vabzdžiai apdulkintojai.

Šio darbo tikslas – nustatyti medunešių bičių gausumą ir jo dinamikos ypatumus dviejų veislių ‘SW Savann’ ir ‘Ural’ vasarinių rapsų pasėliuose. Aukšta oro temperatūra, būdinga visam tyrimų laikotarpiui, suteikė galimybę ištirti augalų ir apdulkintojų sąveiką esant neįprastoms Lietuvai sąlygoms.

Įvertinus vasarinių rapsų dviejų veislių žydėjimo intensyvumą ir bičių lankymąsi jų pasėliuose nustatyta, kad veislės ‘Ural’ pasėliuose žydėjo vidutiniškai 4,6 % daugiau žiedų nei veislės ‘SW Savann’, tačiau bičių tankumas ant veislės ‘Ural’ žiedų buvo vidutiniškai 4 % mažesnis nei ant veislės ‘SW Savann’.

Rapsų žydėjimo intensyvumo mažėjimas koreliavo su bičių tankumo mažėjimu abiejų veislių pasėliuose.

Ekstremalus temperatūros padidėjimas turėjo neigiamos įtakos bičių aktyvumui ant žydinčių augalų. Mažiausiai medunešių bičių buvo aptikta per popietinį stebėjimą (16 val.), kai oro temperatūra pakilo iki +43 °C.

Dėl ekstremalių sąlygų trumpesnis rapsų žydėjimo laikas ir mažesnis apdulkintojų tankumas ant jų žiedų galėjo neigiamai paveikti ir rapsų sėklų derlių.

Reikšminiai žodžiai: vasarinis rapsas, *Brassica napus*, naminės bitės, *Apis mellifera*, apdulkinimas, aplinkos temperatūra.