sciendo

RURAL SUSTAINABILITY RESEARCH
48(343), 2022
ISSN – 2256-0939



Received: 26 September 2022 Revised: 20 October 2022 Accepted: 3 November 2022

Bumblebee pollination activity in a commercial tomato greenhouse during the winter season

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Abstract. Bumblebee activity interaction with solar irradiation, solar irradiation and HPS lighting irradiation, the HPS effect of photoperiod, day temperature and hive density were assessed with tomato fruit weights in a commercial tomato glass greenhouse in 2020-2021. Pollination activity was measured by observing ten randomly chosen cv. 'Beorange' tomato plants and by counting bumblebee bruised tomato flowers. MANOVA was conducted between factors and bee activity, followed by Pearson's correlation. A Mann-Whitney U test was calculated to determine the significance between tomato flower bruising levels and fruit weights, followed by Cliff's delta (*d*). Pollination activity decreased mainly in December and January when solar irradiation decreased to below 110 J cm^{-2} day^{-1}. Bumblebee activity was significantly affected by solar irradiation with HPS lighting ($p<0.001$; $p<0.01$). There was a significant correlation between bumblebee activity and solar irradiation ($r= 0.75$; $p<0.05$), and solar irradiation with HPS lighting ($r= 0.70$; $p<0.05$). There was no correlation between bumblebee activity and fruit weights ($r= -0.20$; $p<0.05$). Bruised flowers had significantly greater fruit weight increases (165.7 g) compared to unbruised flowers (123.4 g) ($d= 0.12$; $p<0.05$). Bee activity rates between 60% to 80% can be concluded as an effective rate for tomato growers. Bumblebees need at least 110-154 J cm^{-2} day^{-1} of solar irradiation to achieve a high pollination activity rate in temperate climate zones during the winter season.

Key words: Bombus terrestris, horticulture, pollinators, tomatoes, greenhouse.

Introduction

Bombus terrestris L. is one of the most common bumblebee species in Europe (Williams & Jepsen, 2020). Most fruit and vegetable crops rely on bumblebee pollination. These pollinators are well known for their ability to pollinate flowers in windy and cold circumstances (Nayak *et al.*, 2020). Because of their ability to fly in low temperatures, bumblebees adapt well to temperate climate zones (Peat *et al.*, 2005). Buzzing ability increases the pollination level and is beneficial for solanaceous crops, such as tomatoes (*Solanum lycopersicum* L.), that require additional vibration (Nayak *et al.*, 2020; Nazer, Kasrawi & Attal, 2003). Although the tomato is a self-pollinating plant, tomato pollination in greenhouses can be negatively affected due to the lack of wind and plant movement. This emphasises the need for bumblebees in greenhouse crop pollination in order to maximise financial benefits (Ahmad *et al.,* 2015; Banda & Paxton, 1991).

Various field and greenhouse studies clearly demonstrate that bumblebee pollinated crops yield high-quality fruit (Ellis *et al.*, 2017; Ercan & Onus, 2003; Kevan *et al.*, 1991; Morandin, Laverty & Kevan, 2001; Yankit *et al.*, 2018). Yankit *et al.* (2018) confirmed that the tomato pollination efficacy of the bumblebees compared to non-bumblebee treatment under protected conditions leads to a higher fruit yield, number per cluster, number of fruits, fruit breadth, weight and length. Researchers also reported, that bumblebee pollinated tomato plants produced rounder fruits (93.33%) than control plants (76.54%). The research of Ahmad *et al.,* (2015) also suggested the use of bumblebees in tomato greenhouses as pollinators, as they significantly increase the fruit's qualitative and quantitative parameters. It is possible that in some cases, the bumblebee visitation rate per flower could also depend on tomato cultivar (Ahmad *et al.,* 2015; Lefebvre & Pierre, 2006) and flower patterns (Hempel

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DOI: 10.2478/plua-2022-0015
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De Ibarra, Langridge, & Vorobyev, 2015). Ahmad *et al.* (2015) in their study found a higher foraging efficacy of *Bombus terrestris* on 'Cherry' tomato cultivar flowers than 'Grandella' cultivar under a greenhouse cultivation system. The opposite study results in commercial tomato greenhouses by Lefebvre & Pierre (2006) revealed no significant differences in bumblebee densities between 'Beefsteak' and 'Cherry' tomatoes. Higher mean densities of available flowers were obtained in 'Cherry' tomatoes (288.57 ± 9.22) than in 'Beefsteak' tomatoes (81.95 ± 5.66) with the average ratio of 3.52. Very few studies are available which describe the importance of flower visitation rates. The study results of the bumblebee foraging behaviour by Morandin, Laverty & Kevan (2001) showed that the highest number of pollen grains from flower stigmas were collected when flower anther cones were bruised in two to three small-to-medium size discolorations, which corresponds up to two bumblebee visits per flower. Zameer *et al.* (2022) concluded, that the best fruit physical parameters were achieved when bumblebees pollinated flowers three times in comparison with hand-held electric vibration and control treatments. Roman & Szczesna (2008) in their study found that bumblebee pollination activity can be also altered by too low (19.6 °C) or too high (24.0 °C) temperature fluctuations in tomato greenhouses. Survilienė, Raudonis & Jankauskienė (2009) studies show that bumblebee activity is effective when pollination is higher than 60% of pollinated tomato flowers. Although, scientists reported a decrease in pollination efficiency after five to six weeks from a new hive introduction in tomato greenhouses. In their study, to avoid a sudden reduction in bumblebee pollination levels, hives were changed one to three weeks earlier.

Bumblebees have been recently introduced as buzz-stimulants for tomato pollination in commercial greenhouses throughout the year in countries with a temperate climatic zone. During winter production, tomatoes as well as bumblebees are exposed to insufficient levels of natural sunlight and it may lower bumblebee pollination levels. Pressman *et al*., (1999) concluded that an increased number of tomato flower pollen grains in the upper clusters are accompanied by the increased temperature and photosynthetic active radiation (PAR) intensity in the spring. Many commercial tomato greenhouses use supplemental lighting, such as high-pressure sodium (HPS) lamps, more often combined with LED light lamps to compensate for the insufficient amount of PAR in winter months, to maintain fruit yield and quality indicators (Olle & Viršile, 2013; Singh *et al*., 2015). HPS lighting has a broad spectral band. HPS lamps mostly emit artificial lighting in red and yellow wavelengths. Compared to natural light sources, HPS blue light output is limited. Recent studies show that blue light is necessary for tomato growth, as it increases the total biomass, yield and the number of fruits, as well as the plant photosynthetic capacity (Kaiser *et al.,* 2019). The lack of blue light spectra can be compensated by artificial LED lighting. In a greenhouse where mini-cucumbers were grown, the vertical spectra of LED lighting under overhead HPS lighting are reported to have a positive influence on the plants physical, quantitative and qualitative parameters (Guo *et al*., 2016). As for tomato plants, study show that the vertical LED regime (top far-red (290 cm from ground) with bottom red LED (160 cm from ground) had the best performance under HPS lighting (Hao *et al.,* 2017). In some research, where the parameters of tomato plant productivity and efficiency of photosynthesis were studied using different artificial lighting sources, HPS had the most total mass of leaves wet weight and dry matter (27.07 ± 1.33 g; 3.15 ± 0.14 g) compared to LED (14.11 ± 0.71 g; 1.37 ± 0.06 g) and fluorescent lighting (25.27 ± 1.26 g, 1.62 ± 0.07 g) (Rakutko S, Rakutko E. & Andrey, 2015).

A study about the effect of decreasing light intensity on bumblebee flight control has been conducted, proving a significant decrease in flight speed and response speed of the bumblebee photoreceptors as the light intensity (lx) decreased (Reber *et al.*, 2015). Roman & Szczesna (2008) results revealed that the flying activity of bumblebees in greenhouses was optimum at a light intensity of 19.6 klx, higher or lower light intensities resulted in the decrease of bumblebee activity. Few studies have described behavioural changes in bumblebee activity with a UV radiation presence in greenhouses (Blacquiere *et al*., 2006; Dyer & Chittka, 2004; González *et al.*, 2004; Morandin, Laverty & Kevan, 2001). The study by Morandin, Laverty & Kevan (2001) reveals that greenhouses equipped with UV light emitting lamps, compared with no UV light treatment, had two times greater impact on the average bumblebee trips per day on tomato flowers. González *et al.* (2004) in their study found that greenhouses not covered in UV light blocking layers retained the original number of bumblebees as in the beginning. However, in other greenhouses where UV-blocking layers were used, the number of bumblebees was reduced between eighteen and nine from the original numbers (36). Blacquiere *et al*. (2006) found significant differences in bumblebee orientation behaviour between glass, polymethylmethacrylate and polycarbonate materials used as greenhouse claddings. The study revealed that when polycarbonate UV light was hardly emitted, bumblebees lost their orientation by not returning to

their hives. In laboratory experiments, opposite results were obtained when Dyer & Chittka (2004) found no significant differences on bumblebee activity with and without a UV light presence. Results were obtained in an environment, where six 40 W light tubes, one 36 W light tube and a UV light emitting screen were used for illumination and diffusion. A UV-blocking filter was used to cover the illumination.

The aim of this research was to determine bumblebee pollination activity relation with various greenhouse environmental factors (day temperature, solar irradiation, solar irradiation with HPS lighting irradiation, the HPS effect of photoperiod hive density) assessing tomato fruit weight during the winter season. No such study has been performed in Latvia until now.

Materials and Methods

Study site

The study and measurements were carried out in the "Getliņi EKO" commercial tomato greenhouse, located at Rumbula, Latvia (GPS coordinates: 56.883705, 24.261453) during the cool-season period in 2020-2021. The first data collection started on 17.09.2020 and ended on 26.03.2021. A total of 18 visits were performed as shown in Fig. 2. Tomatoes were grown in a modern greenhouse with the ceiling height of 5.5 m and an area of 0.73 ha. The roof of the greenhouse is glazed with four millimetre thick glass, which is placed in aluminium profiles and sealed with special rubber. For safety reasons, there are transparent safety nets under the greenhouse roof in case the glass breaks. The greenhouse roof was not covered with any additional film layers or materials which block natural UV light. During the research in the cool season, the greenhouse was heated using converted energy from landfill gas, which comes from biodegradable waste. The greenhouse climate control equipment provided the necessary environment and technological parameters for growing plants as well as meteorological data. The mean temperature of the tomato greenhouse was +21.9 °C (+21.1 °C min, +23.9 °C max.). To compensate for reduced natural light for plants in winter months, on the top of the tomato canopy, HPS supplemental lighting was used with a photoperiod of 17.5 h day^{-1}. For bumblebee pollination activity cultivar 'Beorange' was examined. Previously prepared tomato seedlings were planted in August, in blocks of rock wool (10.0*10.0*6.5 cm). Then, plants were placed in a rock wool plate (100.0*20.0*7.0 cm), hung in planting beds, located 70.0 cm above the ground. A computerized fertilizer mixing and feeding program were used to provide plants with mineral nutrients. The nutrient solution was applied to plants using a drip irrigation system with an accuracy of one mg L^{-1}. Observed tomatoes were harvested twice a week and weighed from the 5th to the 28th cluster. The first tomato cluster began to bloom in August. Clusters bloomed till the end of May when the main growing points were removed.

Description of pesticide use

For the biological control of pests (such as phytophagous mites, thrips, aphids and whiteflies), predatory bugs (*Macrolophus pigmaeus* Wagner), parasitic wasps (*Encarsia formosa* Gahan) and predatory mites (*Neoseiulus californicus* McGregor) from "Biobest Group NV" (Belgium) were used as their natural enemies. A sulphur evaporator "Nivola" from "Royal Brinkman" (Netherlands) was used to control tomato powdery mildew (*Pseudoidium neolycopersici* L. Kiss). Other tomato pests and diseases were not detected during research. No other plant protection products were used.

Description of bumblebee hives

Bumblebee hives from "Biobest Group NV" were used in a commercial tomato greenhouse. Two hives were placed in each position (0.5 m distance from each position to other) on special shelves at a height of 2.4 m from the ground. The shelves were hung from roof construction bars near the glass wall in the middle of the corner of the greenhouse. The bumblebee hives were located four metres from the observed tomato plants. Each hive consisted of a large colony of bumblebee species (*Bombus terrestris* L.), containing 80-110 workers and one queen. Hives were equipped with ventilation and nutrition systems (a pollen tray and sugar substitute). The first three hives were placed in the second week of August. Five hives were placed in the third week of September, but four hives were placed in the third week of December. In the rest of the study period, no more than six hives were placed during data collection. The last colonies of bumblebee hives were placed in the second week of March. Hives were placed and replaced by the "Getliņi" agronomist according to the company policy and guidelines. Hives were used up to six to eight weeks, then old hives were replaced by new ones, following the order of rotation. Throughout the study, a total number of 101 hives were used. All placed bumblebee hive numbers in each data collection day are presented in plot A (Fig. 2.)

Description of bumblebee activity monitoring and methodology

Fully bloomed flowers of tomato clusters were monitored. Bumblebee activity was determined by counting bumblebee bite marks on each individual flower. Bite marks on the tomato flower appear as bruises on the anther cone, on which the jaw imprints of the bumblebee are visible. After three to four days, bite marks in the anther cone were clearly visible

$$\text{Bumblebee activity (\%)} = \frac{\text{Bruised flowers} * 100}{\text{The total number of counted flowers}}$$

Figure 1. Formula represents the bumblebee activity percentage calculation.

as they turned slightly brownish and appeared to be physiologically damaged. For monitoring data collection, the following materials were required: border tape for marking tomato plant (trolley tubes) rows and tomato plants; labels and a marker for marking tomato clusters; different colour threads for flower marking in tomato clusters. Ten tomato plants, located in various rows of tomato plants throughout the greenhouse were selected for the bumblebee activity monitoring. The rows of trolley tubes with the tomato plants were marked with border tape for quick identification. One random tomato plant was selected in each row of trolley tubes. Each selected tomato plant stem was also marked with border tape for quick identification. Numbered labels were attached to the clusters of each labelled tomato plant, listing the formed clusters with flowers sequentially. In each cluster, fully bloomed flowers were tagged with threads of different colours as necessary (blue, red, green, yellow, white and black). The flowers were tagged to separate them from other flowers, and later on to determine whether the fruit had developed (ripened). The activity of bumblebees was expressed as a percentage (%) from bruised flowers and the total number of counted flowers, and calculated according to the following formula (Fig.1.):

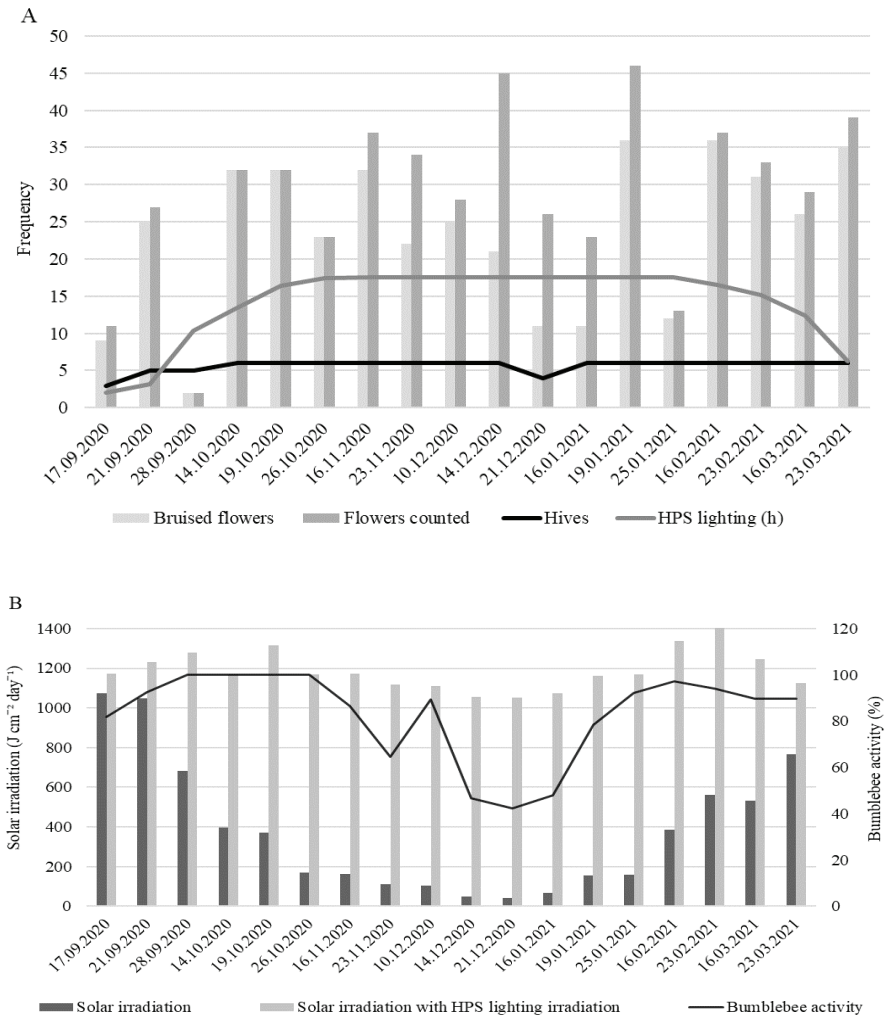


Figure 2. Plot A represents the dynamics of bruised flowers by bumblebees in season 2020-2021. Horizontal bars represent hive density and HPS supplemental lighting hours in each observation. Plot B represents the dynamics of bumblebee activity depending on solar irradiation, as well as the total amount of solar irradiation and HPS lamp lighting irradiation in J cm^{-2} day^{-1}.

Statistical analysis

A multi variance analysis of variance (MANOVA) was conducted to analyse factor interaction between bumblebee activity and solar irradiation, solar irradiation with HPS lighting, the HPS effect of the photoperiod, day temperature and hive density. One-way ANOVA was calculated to find statistically significant differences. The strength of effect size (partial eta-squared, η^2) was calculated and labelled following Field's (2013) recommendations. For statistically significant differences, Pearson's correlation coefficient (r) was calculated to measure the strength of linear associations using a two-tailed significance test. A Mann-Whitney U test was calculated to determine the significance between tomato flower bruising levels and fruit weight. For statistically significant differences, Cliff's delta (*d*) was calculated to determine the effect size (Fox, Weisberg, 2011). The mean, median and standard deviation of fruit weights from tomato flower bruising levels was calculated and shown in a boxplot to summarize observed samples. To determine the significance level, a 95% confidence level was used. All calculations were done using R-Studio software (R Core Team, 2018).

Results

In season 2020-2021, a total of 484 flowers were counted of which 421 flowers were bruised, but 264 fruits were ripened and harvested. A 100% pollination activity rate of bumblebees was registered between September and October with a solar irradiation of 169-681 J cm^{-2} day^{-1} as shown in plot B (Fig. 2). The lowest activity of 42-48% was recorded between December and January, corresponding to a decreased solar irradiation between 47-67 J cm^{-2} day^{-1}. The mean bumblebee activity during the observation period was 86.9%. HPS supplemental lighting with a photoperiod of 17.5 h per day was used between 19.10.2020 and 25.01.2021 as shown in plot A (Fig.2).

During research, an increase of bumblebee activity was detected from 23.11.2020 to 10.12.2020. This could be explained that new colonies of bumblebee hives were replaced by a greenhouse agronomist a few days earlier, and there was higher bumblebee activity on the day of the data collection. However, on 23.11.2020, fewer bruised flowers were recorded in relation to the total number of flowers counted. The number of bruised flowers on 10.12.2020 was higher than on 23.11.2020, but the total number of flowers counted on 10.12.2020 was smaller. As a result, such an increase in bumblebee activity could have occurred.

In the 2020-2021 tomato growing season, the interaction of all studied factors, determined by MANOVA analysis, had a significant influence on bumblebee activity ($p<0.01$) with a large effect size ($\eta^2=0.75$) (Table 1). The results indicate that 75% of the bee activity can be accounted for by the controlled effects of factors in the greenhouse environment. One-way ANOVA showed, that factors, followed by solar irradiation and total irradiation with HPS lighting, had a significant influence on bumble bee activity ($p<0.001$; $p<0.01$) with a large effect size ($\eta^2=0.56$, $\eta^2=0.50$), except for the HPS effect of photoperiod ($p>0.05$), day temperature ($p>0.05$) and hive density ($p>0.05$) with small, medium and small effect size ($\eta^2=0.04$; $\eta^2=0.12$; $\eta^2=0.04$), respectively. Photo periodism from the artificial lighting of HPS had no impact on bumblebee behavioural changes. Bumblebee activity did not increase nor decrease from day temperature and hive density changes throughout the study period. A calculation by Pearson's correlation coefficient revealed a significant strong positive correlation between solar irradiation and bumblebee activity ($r= 0.75$; $p<0.05$), as well as between bumblebee activity and a total irradiation with HPS lighting ($r= 0.70$; $p<0.05$) (Fig. 3). There were linear associations between all variables. The results indicate that as soon as solar irradiation increases, regardless of a HPS lighting presence or absence, bee activity increases significantly.

Pearson's correlation between tomato fruit weight and bumblebee activity revealed a statistically insignificant negative relation ($r= -0.20$; $p>0.05$) (Fig. 4). There was no linear association between the two

Table 1

Comparison of the affecting factors in bumblebee activity in the tomato greenhouse season 2019-2020

Season	Analysis of variance results	Solar irradiation	Solar irradiation and HPS lighting	HPS effect of photoperiod	Day temperature	Hive density	The interaction of all factors
2020 -2021	df	18	18	18	18	18	5
	F	20.255	15.756	0.651	2.088	0.634	7.397
	p-value	0.000	0.001	0.431	0.168	0.438	0.002
	η^2	0.56	0.50	0.04	0.12	0.04	0.75

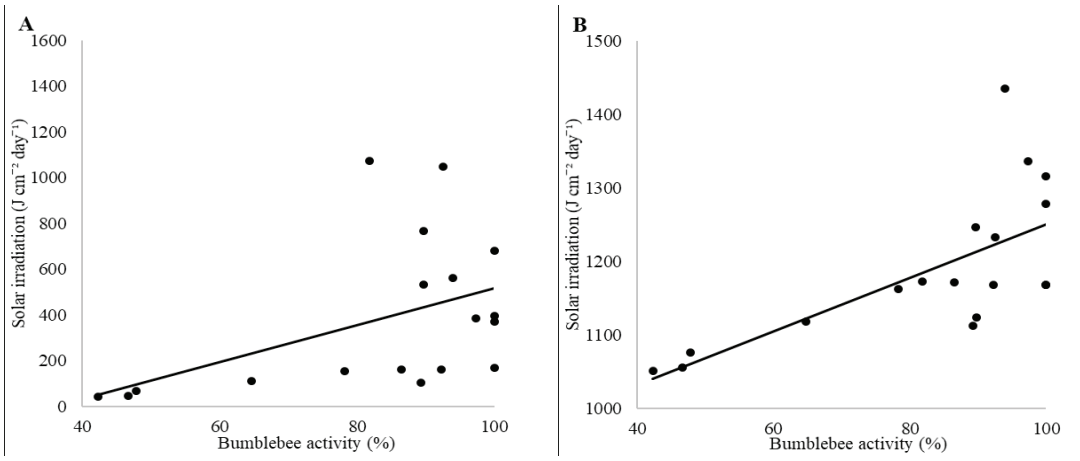


Figure 3. Plot A represents a significant linear relationship between solar irradiation and bumblebee activity in the 2020-2021 tomato growing season, calculated by Pearson's correlation. Plot B represents a significant linear relationship between solar irradiation with HPS supplemental lighting irradiation and bumblebee activity in the 2020-2021 tomato growing season.

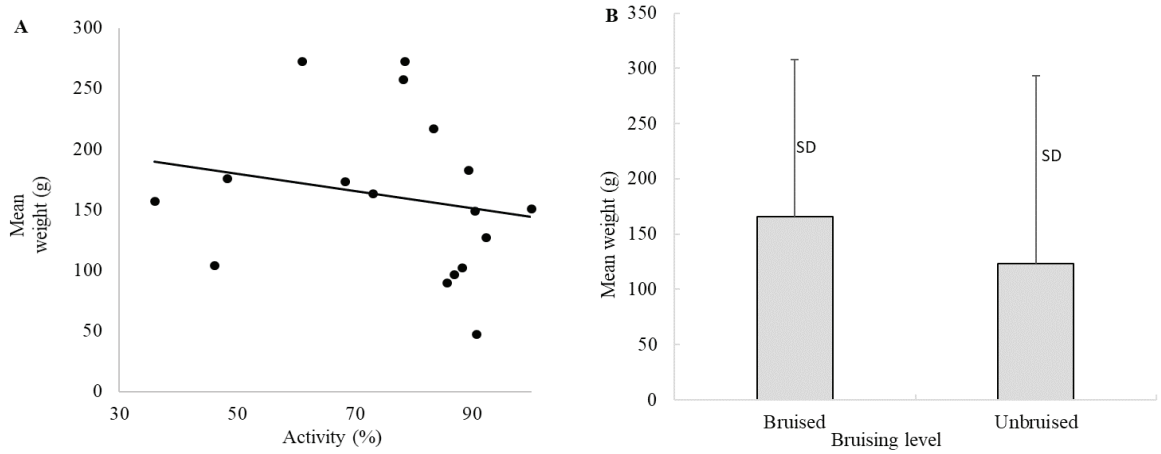


Figure 4. Plot A represents an insignificant relationship between the mean number of tomato fruit weight (n=433) and bumblebee activity in the 2020-2021 tomato growing season, calculated by Pearson's correlation. Plot B represents the relationship between tomato fruit weight and bumblebee bruising level of tomato flowers in the 2020-2021 tomato growing season.

variables. Observations showed that pollination had no influence on tomato fruit weight changes. The observed bumblebee activity showed that the highest fruit weight increase was obtained when the activity rates were between 60% and 80%.

A Mann-Whitney U test showed that there was a significant difference in fruit weight changes (U=9394; p=0.011, α=0.05) between the group of tomatoes whose flowers were bruised by bumblebees and the group of tomatoes whose flowers were unbruised. The observed effect size was small (*d*=0.12), indicating that the magnitude of the differences between bruised and unbruised flowers were small, but significant. Calculations showed that flowers, which received bruising treatment, developed 64% of the tomato fruits compared to flowers, which did not receive bumblebee bruising and developed only 41% of the tomato fruits (out of 100%). The median fruit weight was 191 g for the bruised tomato flowers compared to zero grams for those flowers not receiving the bumblebee bruises suggesting that bumblebee bruising is more effective. The observed mean fruit weight (g) and standard deviation showed that fruits from flower bruising had greater weight increase (165.7 g) ($\bar{x}$= ± 142.5 g SD, n=370) in comparison to fruits whose flowers were unbruised (123.4 g) ($\bar{x}$= ± 169.4 g SD, n=63) (Fig. 3.).

Discussion

All factor interaction with bumblebee activity showed that 75% of the bee activity can be accounted for by the controlled effects of factors in the greenhouse environment, suggesting that the uncontrolled factors

could be the cause of fertilizers or even pesticides (Ceuppens, *et al*., 2015; Mommaert & Smagghe, 2011; Survilienė, Raudonis & Jankauskienė, 2009). Solar irradiation and solar irradiation with HPS lighting irradiation had a significant impact on bumblebee activity ($p<0.001$; $p<0.01$) in season 2020-2021 in comparison only with the HPS effect of photoperiod ($p>0.05$), day temperature ($p>0.05$) and hive density ($p>0.05$). The results by Pearson's correlation clearly showed that the presence (solar irradiation output) of HPS lighting was important for bumblebee activity in a greenhouse environment. Although, during December and January when solar irradiation was the lowest, between 47-67 J cm^{-2} day^{-1}, the presence of HPS supplemental lighting was unable to provide sufficient bumblebee activity. The results showed that bumblebee activity, mainly in December and January, had a tendency to decrease by more than half (42-48%) as solar irradiation decreased below 110 J cm^{-2} day^{-1}. According to Survilienė, Raudonis & Jankauskienė (2009), the bumblebee pollination activity is effective when 60% of tomato flowers are pollinated, and new hive placement is suggested after one to three weeks to maintain the pollination activity. The results from the data observations showed that the mean activity of 86.9% was reached. Few observations showed that bumblebee activity of 60% was not reached when solar irradiation decreased below 104 J cm^{-2} day^{-1}. There was an exception when bumblebee activity increased up to 89% when solar irradiation reached 104 J cm^{-2} day^{-1}, but it is unclear why pollination activity of 89% was higher in 10.12.2020, than it was in 31.11.2020 when activity of 65% was reached at a solar irradiation of 110 J cm^{-2} day^{-1}. It is possible, that new colonies of bumblebee hives were replaced a few days earlier and there was higher bumblebee activity on the day of the data collection.

From the data observations, it can be considered that bumblebees need at least 110 J cm^{-2} day^{-1} of solar irradiation to achieve activity of more than 60%. Changes in bumblebee activity could be also explained by increased or decreased hive density and the frequency of the hive colony replacement. However, in this study, no more than six hives were placed or replaced by new hives every second week throughout the observation period and hive density had no significant influence on bumblebee activity ($p>0.05$). Study by Pressman *et al*., (1999) showed that bumblebees can't pollinate properly without sufficient solar irradiation, indicating that the solar irradiation and the range of its light wavelengths has a significant impact on bumblebee orientation in the environment, but it is one of the few influencing factors. Although solar irradiation combined with HPS lighting had a significant influence on bumblebee activity, it wasn't enough to archive high pollination activity in December and January. Decreased bumblebee activity in winter months could be associated with the low light source (as well as an insufficient UV light source) in the early morning hours when colony traffic and foraging activity reaches the highest peak (Ahmad *et al.,* 2015; Kwon & Saeed, 2003). In this study, the duration of HPS lighting had no significant impact on bee activity ($p>0.05$). Roman & Szczesna (2008) and Reber *et al.* (2015) tested different light intensities on bumblebee flying activity, proving a significant decrease in flight speed and response speed of the bumblebee photoreceptors as light decreased. Behavioural changes in bumblebee activity can be altered by the presence of UV light, which is essential for bumblebee orientation and flower searching time (Blacquiere *et al*., 2006; Dyer & Chittka, 2004; González *et al.*, 2004; Morandin, Laverty & Kevan, 2001). One of the possibilities to increase bumblebee activity in winter months could be the use of UV emitting light sources to intensify bumblebee orientation (Morandin, Laverty & Kevan, 2001). According to González *et al.* (2004) and Blacquiere *et al*. (2006) studies, UV reflective materials should not be used in greenhouses, as it may decrease the number of bumblebees remaining in the hives (González *et al.*, 2004). To compensate for low bumblebee activity in late winter months, a hand-held electric vibration treatment is considerable (Kwon & Saeed, 2003) as it shows promising results when used for at least two minutes (Zameer *et al.,* 2022).

In this study, the observations showed that highest tomato fruit weight increase was reached between 60% and 80% of the bumblebee activity rate, which is effective according to Survilienė, Raudonis & Jankauskienė (2009), but Pearson's correlation results between two variables showed no linear relationship ($r= -0.20$; $p>0.05$). The results could be explained by bee behavioural changes in winter months (Dyer & Chittka, 2004) and the fact that the tomato greenhouse is located in a temperate climate zone where the natural light source is low in December and January. Perhaps, a comparison of tomato fruit shape, seed count, diameter and roundness between different pollination levels would show different results, but no such fruit physical parameters were investigated due to different project tasks.

Tomato flowers were divided into two groups (bruised and non-bruised, regardless of bumblebee visitation rate) to determine bumblebee activity. A significant difference by Mann-Whitney U test was found in fruit weight changes ($U=9394$; $p=0.011$, $\alpha=0.05$) between two groups. Although the effect size was small ($d=0.12$) due to the effect of different factor interaction, findings showed that bumblebee bruising

were more effective than non-bruising treatment. Moreover, it was found between two groups, that bruised flowers developed 64% of the fruits and were heavier compared to non-bruised flowers developing 41% of the fruits. Morandin, Laverty & Kevan (2001) in their study found that the highest number of pollen grains from flower stigmas were collected when flowers were bruised in two to three, small to medium size, discolorations (equals one to two bee visits), but Zameer *et al.* (2022) in their study found that tomato flower bruised three times had the highest fruit physical parameters. It is possible, that collecting pollen grains from flowers of different bruisings would give more accurate results to predict bumblebee activity. Additional measurements such as colony flying rate from bumblebee hives at different time periods, comparison between fruit physical parameters and bruisings are required to determine bumblebee activity in a long-term for further studies.

Conclusions

1. Solar irradiation and solar irradiation with HPS supplemental lighting irradiation had a significant impact on bumblebee activity in the winter tomato growing season ($p<0.001$; $p<0.05$), compared to the HPS effect of photoperiod, day temperature and hive density ($p>0.05$; $p>0.05$; $p>0.05$), respectively. Findings suggest that more observations should be conducted over a longer term to compare the results between seasons.

2. Bumblebee activity, mainly in December and January, had a tendency to decrease when solar irradiation decreased below 110 J cm^{-2} day^{-1}. It is concluded that bumblebees need at least 110-154 J cm^{-2} day^{-1} of solar irradiation output to achieve high pollination activity in a temperate climate zone during the winter season.

3. In the study, the highest tomato fruit weight increase was reached between 60% and 80%, and can be concluded as an effective activity rate for tomato growers. Bumblebee bruising treatments on flowers were significantly more effective on tomato fruit weight changes compared with non-bruising treatments.

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Acknowledgment

The study was carried out during the Nr. 18-00-A01612-000017 research project "Determining of the optimal growth conditions for vegetables using hydroponics cultivation method with artificial and natural lighting", based on "ELFLA" Rural development 2014-2020 activity for Operational Groups. I am thankful to the reviewers for their time and suggestions for improving the manuscript.