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Flight activity of aphids in Serbia: Investigation by water traps placed in sugar beet fields

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Abstract: Plant viruses, and aphids as their vectors, are limiting factors in sugar beet production. Viral plant diseases are currently impossible to treat, but knowing the flight patterns of aphids can help reduce the number of potential virus vectors. Monitoring of aphid flight activities in sugar beet fields was done using yellow water traps from April to the end of November. During the two years of investigation, a total of 5 514 specimens from 75 different taxa were collected. All localities recorded the highest number of individuals at the end of May/beginning of June. This is the period when sugar beet develops intensively, so the risk of virus infection is the highest. The most numerous species were *Aphis fabae* Scopoli, *Aphis spiraecola* Patch., *Phyllaphis fagi* (L.), *Myzus persicae* (Sulzer), *Rhopalosiphum padi* (L.), *Sitobion avenae* (Fabr.), *Acyrthosiphon pisum* (Haris) and *Therioaphis trifolii* (Monell). The most important vectors are *A. fabae*, *A. spiraecola* and *M. persicae*. *Aphis fabae* is a species that feeds on sugar beet and causes significant damage by feeding and its vector activity. *Myzus persicae* was the most abundant in autumn, *A. spiraecola* was present throughout the whole flight-monitoring period. Among the caught aphids, twelve species alien to Europe were collected.

Keywords: Aphididae; *Beta vulgaris* v. *altissima*; vectors of viruses; invasive species

Sugar beet (*Beta vulgaris* v. *altissima* Ross.) is Europe's main crop for sugar production. The production of sugar beet is affected by many aphid species that can reduce the yield and quality of the plants by sucking the sap and by vectoring sugar beet viruses (Qi et al. 2004). Sugar beet is host to ten cosmopolitan aphid species, mostly extremely polyphagous, such as *Aphis fabae* Scopoli, *Myzus persicae* (Sulzer), *Macrosiphum euphorbiae* (Thomas), *Aphis gossypii* Glover, *Aphis nasturtii* Kaltenbach and *Aulacorthum solani* (Kaltenbach) (Blackman & Eastop 2022). The two main aphid species colonizing sugar beet worldwide are *A. fabae* and *M. persicae*, which form large colonies and can be sustained throughout the whole season on sugar beet plants (Fernandez-Quintanilla et al. 2002; Razmjou & Fallahi 2009). *Myzus persicae* is a highly polyphagous species and a significant vector of plant viruses. However, all these species are vectors of sugar beet viruses, and the damages caused by viral infection

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are often much greater than those caused by feeding. There are four viruses transmitted by aphids that damage the beet crop in Europe: beet yellows virus (BYV), beet mild yellowing virus (BMYV), beet chlorosis virus (BChV), and beet mosaic virus (BtMV) (Hossain et al. 2021). BYV is transmitted semi-persistently by more than 20 aphids, *M. persicae* and *A. fabae* being the main vectors. *M. persicae* and *M. euphorbiae* persistently transmit BMYV and BChV. BtMV is transmitted in a nonpersistent way; the main vectors are *M. persicae* and *A. fabae*, but also *Myzus ascalonicus* Doncaster, *M. euphorbiae*, *Acyrthosiphon pisum* (Haris), *Metopolophium dirhodum* (Walker) and *Rhopalosiphum padi* (L.) (Hossain et al. 2021). Viral infection can decrease sugar yield by up to 25% (Clover et al. 1999) or even up to 47% (Smith & Hallsworth 1990), depending on the virus species and time of infection. The importance of monitoring aphid species as potential vectors of viruses in the production of sugar beet has been growing in recent years. Climate change favours aphid overwintering due to milder winters, and a more intensive spreading of viruses across sugar beet fields is expected in the near future. At the same time, globalization and climate change favour the establishment of new species (Coeur d'Acier et al. 2010). In the last 12 years, 10 new invasive aphid species have been detected in Serbia (Petrović-Obradović et al. 2010; Petrović-Obradović et al. 2018; Petrović-Obradović 2021), one being caught by yellow water trap (Vučetić et al. 2014).

Fields growing sugar beet in Europe and Serbia have steadily declined since 2017. In 2020, sugar beet was grown on about 2.89 million ha in Europe (a 15% decrease compared to 2017) and on 37 400 ha in Serbia (a 30% decrease compared to 2017), according to FAOSTAT data (https://www.fao.org/faostat/en/#data). In 2018, the European Commission banned the use of three neonicotinoids (clothianidin, imidacloprid, thiamethoxam), except for use in greenhouses, because of their harmful effect on wild bees and honeybees (European Commission 2018a, b, c). These neonicotinoid pesticides are important for sugar beet production because they are used to control aphids that spread virus yellows. Implementing these regulations has led to a significant decrease in France's sugar beet yield in 2020 (USDA 2020).

The main goals of this study were to monitor the presence of winged aphids in sugar beet crops, including an inventory of aphid species flying over sugar beet fields and aphid flight activity, recording the most abundant species and eventually trapping new alien species. Establishing the time of flying and the numerousness of potential vectors of beet viruses are also very important goals.

MATERIAL AND METHODS

Aphid monitoring was done on two experimental fields of the Institute of Field and Vegetable Crops, Novi Sad, Vojvodina, northern Serbia, at Rimski Šančevi [Rimski Šančevi 1 – RŠ1, 2 ha (N 45.332886, E 19.830680) and Rimski Šančevi 2 – RŠ2, 6 ha (N 45.323923, E 19.817748)], during 2019 and 2020 and on a field (12 ha) of sugar beet in Temerin (near Novi Sad, N 45.411311, E 19.916631) in 2019. At each locality, two yellow water traps (30 × 30 cm) were set up mid-April, when average temperatures were above 10 °C (www.hidmet.gov.rs/data/meteo_godisnjaci/ 2019; www.hidmet.gov.rs/data/meteo_godisnjaci/ 2020), immediately after the emergence of sugar beet. They were in place until the end of vegetation (mid-November). Samples with aphids were taken once per week and kept in tubes with 70% alcohol. Aphids were taken to the Laboratory of Entomology at the Faculty of Agriculture in Belgrade. They were identified using a stereoscopic microscope and keys for identifying winged aphids (Taylor 1984; Jacky & Bouchery 1988; Remaudière & Seco Fernandes 1990; Rongai & Cerato 2001). Some individuals were mounted on slides using standard methods (Eastop & van Emden 1972) and compared with the aphid collection of the authors.

RESULTS

Presence of aphid species

During the two study years, at the three localities (Rimski Šančevi 1 – RŠ1, Rimski Šančevi 2 – RŠ2 and Temerin), a total of 5 514 specimens of aphids (Hemiptera: Aphididae) were trapped. Seventy-five different aphid taxa were identified: 70 in 2019 and 50 in 2020. Altogether, 47 taxa were identified at the species level, 27 at the genus level, and one at the subfamily level (Table 1). In some cases, aphids could not be identified at the species level but only as genera or subfamilies. This is because

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Table 1. Aphid taxa, the absolute number and relative contribution to the total number of caught aphids per year

Identified aphid taxa	2019		2020		Identified aphid taxa	2019		2020	
	n	%	*n*	%		*n*	%	*n*	%
Acyrthosiphon pisum	32	0.87	31	1.67	*Macrosiphoniella* spp.	2	0.05	0	0
Acyrthosiphon cyparissiae	2	0.05	0	0	*Macrosiphum albifrons*	1	0.03	0	0
Amphorophora rubi	23	0.63	2	0.65	*Macrosiphum euphorbiae*	10	0.3	7	0.37
Anoecia spp.	4	0.11	2	0.11	*Macrosiphum rosae*	7	0.20	3	0.16
Anoecia corni	22	0.60	29	1.56	*Metopolophium dirhodum*	49	1.33	4	0.21
Aphis spp.	693	18.90	59	3.17	*Myzus* spp.	18	0.50	4	0.21
Aphis craccivora	50	1.36	7	0.38	*Myzus ascalonicus*	3	0.08	0	0
Aphis gossypii	55	1.50	38	2.04	*Myzus cerasi*	3	0.08	0	0
Aphis fabae	435	11.86	372	20.03	*Myzus persicae*	121	3.30	506	27.24
Aphis pomi/spiraecola	479	13.06	66	3.55	Myzocallidinae	53	1.44	14	0.75
Aphis sambuci	0	0	12	0.64	*Myzocallis castanicola*	2	0.05	0	0
Atheroides serulatus	1	0.03	1	0.05	*Myzocallis komareki*	1	0.03	0	0
Appendiseta robiniae	1	0.03	0	0	*Nasonovia ribisnigri*	3	0.08	0	0
Aulacorthum solani	20	0.54	14	0.75	*Ovatus inulae*	0	0	1	0.05
Brachycaudus helichrysi	22	0.60	23	1.24	*Pemphigus* spp.	89	2.42	41	2.21
Brachycaudus spp.	20	0.54	16	0.86	*Periphyllus* spp.	3	0.08	1	0.05
Brevicorynae brassicae	13	0.35	2	0.38	*Phorodon humuli*	58	1.58	13	0.70
Capitophorus spp.	11	0.30	11	0.59	*Phyllaphis fagi*	684	18.65	45	2.42
Cavariella spp.	2	0.05	6	0.32	*Rhopalosiphum padi*	146	3.98	49	2.64
Cinara spp.	3	0.08	0	0	*Rhopalosiphum maidis*	14	0.38	6	0.32
Chaitophorus populialba	1	0.03	0	0	*Rhopalosiphoninus latysiphon*	2	0.05	1	0.05
Chaitophorus spp.	4	0.11	12	0.64	*Semiaphis* spp.	2	0.05	1	0.05
Cryptomyzus spp.	10	0.3	0	0	*Schizaphis graminum*	10	0.3	0	0
Diuraphis noxia	2	0.05	1	0.05	*Sipha* spp.	4	0.11	0	0
Dysaphis spp.	6	0.16	2	0.11	*Sitobion avenae*	13	0.351	17	0.91
Drepanosiphum spp.	3	0.08	0	0	*Sitobion fragariae*	2	0.051	0	0
Drepanosiphum aceris	1	0.03	0	0	*Smynthurodes betae*	0	0	1	0.05
Drepanosiphum platanoides	19	0.52	0	0	*Subsaltusaphis* spp.	1	0.03	4	0.21
Eriosoma spp.	36	0.98	1	0.05	*Therioaphis trifolii*	41	1.12	70	3.77
Eucallipterus tiliae	3	0.08	1	0.05	*Therioaphis* spp.	10	0.27	0	0
Euceraphis sp.	1	0.03	0	0	*Tetraneura* spp.	33	0.90	15	0.81
Euceraphis betulae	0	0	18	4.362	*Tinocallis* sp.	1	0.023	0	0
Forda sp.	1	0.03	0	0	*Tinocallis saltans*	1	0.03	1	0.05
Hayhurstia atriplicis	1	0.03	0	0	*Tinocallis takachihoensis*	0	0	1	0.05
Hyadaphis spp.	110	3.00	16	0.86	*Trichosiphonaphis polygonifoliae*	2	0.05	1	0.05
Hyalopterus pruni	39	1.06	100	5.38	*Tuberculatus* spp.	7	0.19	0	0
Hyperomyzus spp.	5	0.13	24	1.29	*Uroleucon* spp.	6	0.16	13	0.70
Lipaphis erysimi	1	0.03	0	0	Aphididae	134	3.65	94	5.06

some insects had been badly damaged because of high temperatures during the summer months, or because the winged females of similar species are too morphologically alike, making identification impossible without knowing the host plants or other forms (apterous, larvae, sexual forms). Some genera (*Aphis*, *Cinara*, *Chaitophorus*, *Uroleucon*, *Macrosiphoniella*, *Brachycaudus*, *Tetraneura*, *Pemphigus*) are especially difficult to identify when only winged forms are known. In 2019, the most

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abundant species was *Phyllaphis fagi* (L.), with 684 collected specimens, followed by the taxon *Aphis pomi/spiraecola* with 479 collected specimens, and *A. fabae*, with 435. *Aphis pomi* De Geer and *A. spiraecola* are morphologically very similar, and their differentiation is difficult using only winged forms. For this reason, they share an entry in Table 1. Sugar beet is one of the preferred hosts of *A. fabae*, and the presence of this species in high numbers in traps is to be expected. *Aphis* spp. were numerous in 2019, with 693 specimens, but fewer were caught in 2020 (only 59).

In 2020, *A. fabae* provided many specimens (372). The number of *A. pomi/spiraecola* and *P. fagi* specimens decreased (66 and 45, respectively), while the number of *M. persicae* increased (506) and contributed to more than 27% of all collected specimens in 2020.

Cereal and maize aphids [*Anoecia corni* Fabricus, *S. avenae*, *R. padi*, *Rhopalosiphum maidis* (Fitch), *Schisaphis graminum* (Rondani), *M. dirhodum*, *Diuraphis noxia* (Kurdjumov)] were regularly found in the traps in both years. In both years, alfalfa aphids, *A. pisum* and *Therioaphis trifolii* (Monell), were found in significant numbers, 63 and 111.

Aphid flight activity began in both years in the third half of April, when the average temperature reached 15 °C.

Distribution of aphids during the season

Distribution of aphids during the season at the RŠ1 locality. At the RŠ1 locality, the first specimens were collected in the first week of sampling (April 22 in 2019 and April 30 in 2020). In 2019, the first samples contained seven individuals, while in 2020 sampling started with 78 collected individuals. Peaks were reached at the end of May when the average number of aphids per trap was more than 200 in 2019 and more than 100 in 2020 (Figure 1). Aphids were in low numbers during the summer, and the next peak was at the end of October. A higher number of individuals in 2019 was due to the large number of *P. fagi*. The following year the most abundant species in this period was *M. persicae*. In 2019 the most abundant vector species was *A. fabae*. It was found at locality RŠ1 in a large number at the beginning of the season when plants are most susceptible to viral infection (Figure 2). The following year, *M. persicae* was the predominant vector species, appearing in large numbers throughout May (Figure 3).

Distribution of aphids during the season at the RŠ2 locality. In the 2019 season, the first specimens were collected on April 22 (seven specimens). A peak was reached at the end of May. The first 12 individuals were collected the following year on May 7, and the peak was reached at the beginning of June. In both years, the average number of specimens at the peaks was between 60 and 70 (Figure 4). In 2019, the most abundant species were *A. fabae* and *A. pomi/spiraecola*. The most effective virus vectors occurred in both years in late May when plants were very susceptible to virus infec-

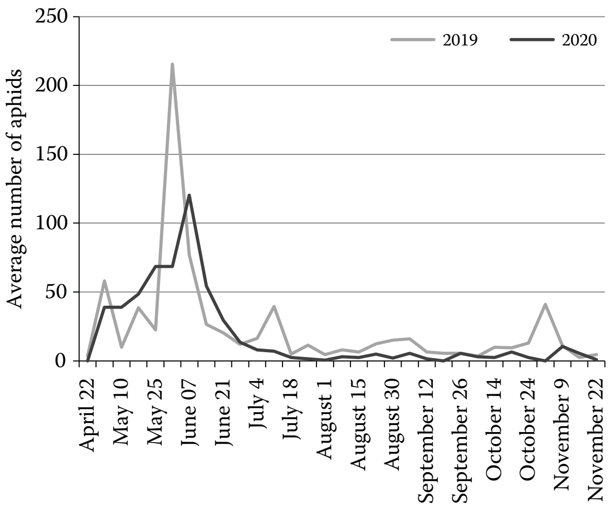

Figure 1. Abundance of aphids in yellow water traps in 2019 and 2020 at locality Rimski Šančevi 1

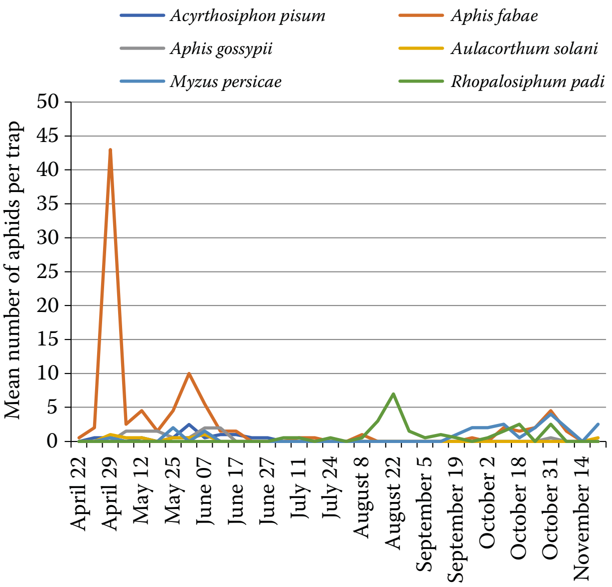

Figure 2. Flight activity of vectors of viruses at locality Rimski Šančevi 1 during 2019

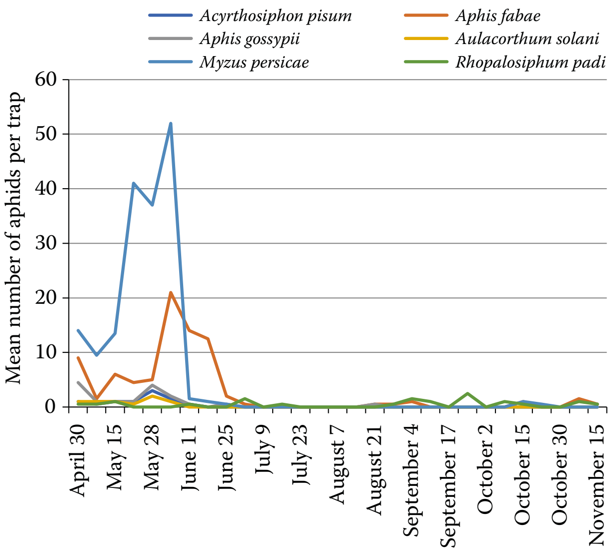

Figure 3. Flight activity of vectors of viruses at locality Rimski Šančevi 1 during 2020

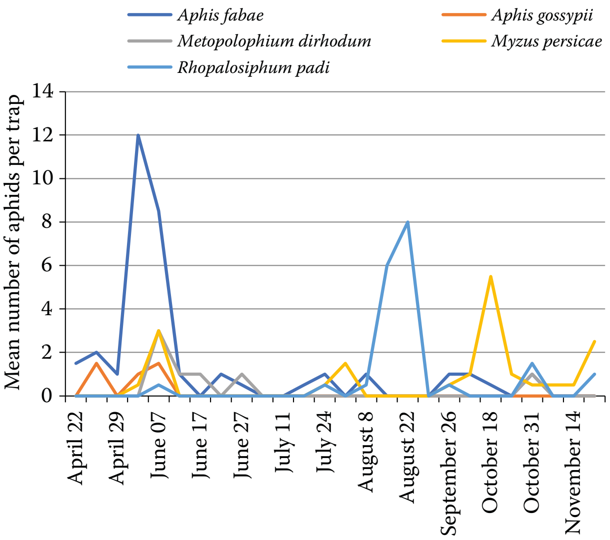

Figure 5. Flight activity of vectors of viruses at locality Rimski Šančevi 2 during 2019

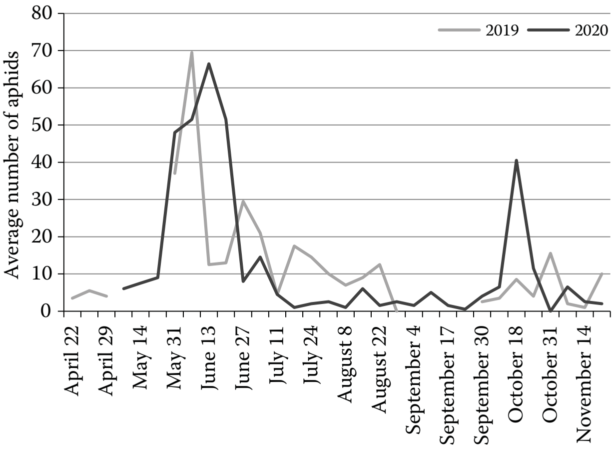

Figure 4. Abundance of aphids in yellow water traps in 2019 and 2020 at locality Rimski Šančevi 2

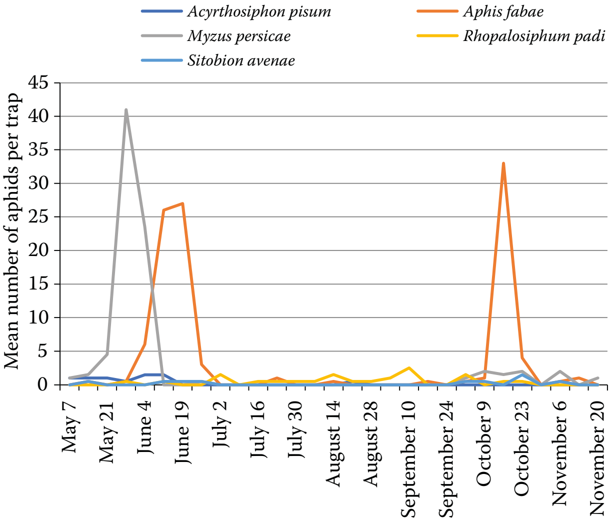

Figure 6. Flight activity of vectors of viruses at locality Rimski Šančevi 2 during 2020

tion. In 2019 it was *A. fabae* (Figure 5), and the following year it was *M. persicae* (Figure 6).

Distribution of aphids during the season at the Temerin locality. The flight activity of aphids at Temerin was monitored in 2019. The first eighteen individuals were collected in the first week of monitoring (April 24). Aphids were numerous at the beginning of June, when the average number of aphids per trap was more than 120 (Figure 7). Aphids were present during the whole season. Six aphid species were found only there: *Acyrthosiphon cyparissiae* (Koch), *Appendiseta robiniae* (Gillette), *Drepanosiphum aceris* Koch, *Hayhurstia atriplicis* (L.), *Myzocallis castanicola* Baker, *Myzocallis komareki* (Pašek). Vectors of viruses such as *A. fabae* and *M. persicae* were present in late May but were most numerous at the end of the season, in mid-October (Figure 8).

Invasive species

During this research, twelve aphid species alien to Europe were collected: *A. gossypii*, *A. spiraecola*, *A. robiniae*, *Macrosiphum albifrons* Essig, *M. euphorbiae*, *M. ascalonicus*, *M. persicae*, *Rhopalosiphoninus latysiphon* (Davidson), *R. maidis*, *Tinocallis saltans* (Nevsky), *Tinocallis takachihoensis* (Higuchi.) and *Trichosiphonaphis polygonifoliae* (Shinji). Some have been present and widely distributed for decades, such as *A. gossypii*, *M. euphorbiae*, *M. ascalonicus*, *M. persicae*, and

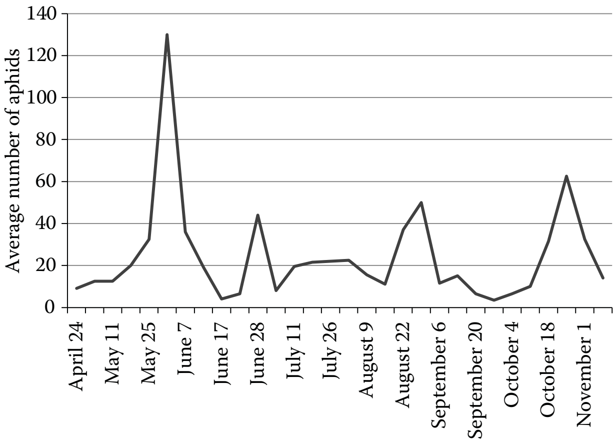

Figure 7. Abundance of aphids in yellow water traps in 2019 at locality Temerin

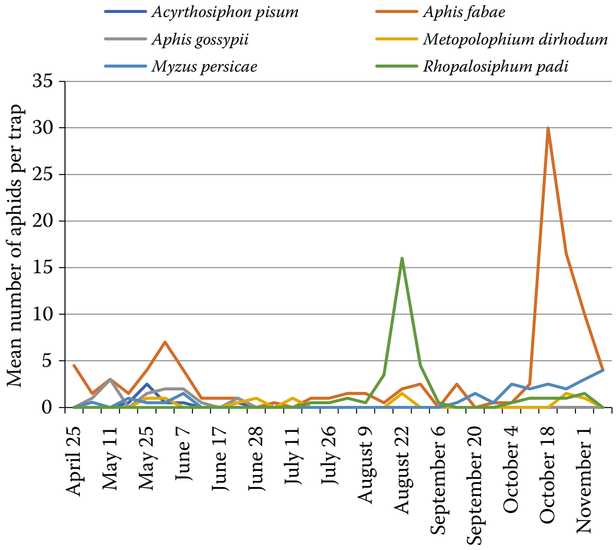

Figure 8. Flight activity of vectors of viruses at locality Temerin during 2019

R. maidis. Aphis spiraecola was found at all localities throughout the season. Others, such as *A. robiniae, M. albifrons, T. saltans, T. takachihoensis* and *T. polygonifoliae,* were found in low numbers. *Rhopalosiphoninus latysiphon* is new for the fauna of Serbia. Additionally, these are the second findings of *M. albifrons, T. saltans, T. takachihoensis* and *T. polygonifoliae* in Serbia.

DISCUSSION

During the two years of investigation, 5 514 specimens from 75 different aphid taxa were collected. The most numerous species were *A. fabae, A. pomi/spiraecola, M. persicae* and *P. fagi*. Both *A. fabae* and *M. persicae* are severe pests of sugar beet, causing crop losses (Fernandez-Quintanilla et al. 2002), and their presence in high numbers was expected. As mentioned above, *A. pomi* is morphologically very similar to *A. spiraecola*, and we analyzed the trapped specimens together. However, *A. pomi* is a pest of apples; it has a short cycle, and winged females are usually not present after July because they lay their eggs very early. On the other hand, *A. spiraecola* is a very polyphagous species (Holman 2009) and produces many winged females during summer and autumn. It has been known in Serbia for only 13 years (Petrović-Obradović et al. 2009). Since then, it has been caught in yellow water traps placed in potato, alfalfa, and oilseed pumpkin fields (Vučetić et al. 2013; Jovičić et al. 2017).

The woolly beech aphid *Phyllaphis fagi* was among the most numerous species, especially in 2019. It is a pest of European beech (*Fagus silvatica* L.) and is often found in yellow water traps in Serbia (Vučetić et al. 2013). Beech trees are found near the experimental sites, resulting in many specimens. Also, *P. fagi* produces a lot of winged forms during spring and summer that fly about looking for another beech tree.

Some of the collected species (*A. cyparissiae, Atheroides serulatus, Appendiseta robiniae, Lipaphis erysimi, Myzocallis castanicola, Myzocallis komareki, Tinocallis saltans, T. takachihoensis*) were found in just one year or only one locality.

The invasive aphid *Rhopalosiphoninus latysiphon* has been found for the first time in Serbia (Petrović-Obradović et al. 2022). This is the second finding of *M. albifrons* in Serbia, found only in yellow water traps in potato fields (Vučetić et al. 2014). *Tinocallis saltans* was recently found on *Ulmus* in Belgrade. This is also a second finding for *Tinocallis takachihoensis* and *Trichosiphonaphis polygonifoliae,* known from only one locality (Belgrade).

If we consider the presence of aphids as vectors of viruses, we can conclude that the risk of infection is relatively high. Potential vectors of sugar beet viruses were found at all localities, but their abundance and distribution over time differed. Important vectors such as *A. pisum, A. fabae, A. gossypii, Aphis sambuci* L., *A. solani, B. brassicae, H. atriplicis, M. dirhodum, M. euphorbiae, M. persicae, R. padi* and *S. avenae* were registered during the investigation, but their abundance differed depending on the locality and year. The intensity of the diseases depends on the density of the aphid populations and the infection source. *Myzus persicae* and *A. fabae* are the most effective vectors and their presence was the highest at the beginning of the season, which is the most sensitive stage of plant development. Early infestation by many aphids that have settled on young sugar beet plants can cause severe epidemics (Van der Werf et al. 1992). Old plants are less susceptible to aphid infestation and virus transmission (Williams 1995). The danger of an early and strong attack is greater when weather conditions favor the propagation of aphids (mild winters and warm weather in the spring). In such conditions, aphids quickly attack sugar beet plants in the spring, where they continue to feed and reproduce. Also, Williams (1995) has shown that *M. persicae* develops much better on infected than on healthy plants, which is important in spreading viruses in the field. *Myzus persicae* is the most effective vector of BMYV and BChV (Kozłowska-Makulska et al. 2009). *Myzus persicae* and *A. fabae* and are the most effective BYV vectors (Limburg et al. 1997). During the research, the first plants with BYV symptoms were observed at the beginning of June 2019 on the experimental fields, and later the presence of BYV was confirmed (unpublished data). This virus causes significant damage in some European countries: in the case of early infection, sugar yield could be reduced by 47% (Smith & Hallsworth 1990), but such damages have not been recorded so far in Serbia. Also, BtMV was registered at the RŠ2 locality in the first year of investigation (unpublished data). This virus is transmissible by more than 28 aphid species (Kennedy et al. 1962) but *M. persicae* and *A. fabae* are the principal vectors in the field, as well as *A. spiraecola* (FERA – UK). Transmission is of the nonpersistent type. Information on the damage caused by BtMV is rare in the literature, but it is generally accepted that this disease has little impact on the yield of sugar beet (Watson & Watson 1953). The intensity of the diseases depends on the density of the populations of aphids and the infection source.

The role of *A. spiraecola* in the spreading of plant viruses is known. It is a very effective vector of more than 20 plant viruses, including cucumber mosaic virus, plum pox virus, potato virus Y, alfalfa mosaic virus, watermelon mosaic virus, zucchini yellow mosaic virus and beet mosaic virus (www.InfluentialPoints.com, https://planthealthportal.defra.gov.uk). During the last several decades, it has widened its distribution in central European countries and is found on many plants (Holman 2009). Climate change affects the environment through changes in mean temperature, precipitation level, and desertification in some areas. The changes are also evident in insects, plants, pathogens, and their interaction. Higher mean temperatures in the growing area of Serbia have created favourable conditions for *A. spiraecola*, a pest of warmer climates.

Monitoring aphid flight activity has provided essential data on the aphid species flying over sugar beet fields and the most abundant species during their peak activity. We also obtained data on the number of potential beet virus vectors and the time of most significant risk for plant infection by viruses. One new invasive species in Serbia was caught. All these data on aphids as virus vectors can help improve crop protection systems.

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