doi:10.14720/aas.2020.115.2.1280 Original research article / izvirni znanstveni članek

Relationship between *Aphis spiraecola* Patch, 1914 (Hemiptera: Aphididae) and citrus foliar minerals

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Received September 27, 2019; accepted May 3, 2020.
Delo je prispelo 27. septembra 2019, sprejeto 03. maja 2020.

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Abstract: Spring and autumn flushes are generally the most infested periods by citrus aphids. Nevertheless, the role of citrus foliar minerals on aphids is not clear. Thus, this paper aims to study the correlation between certain minerals and the infestation degree of citrus varieties by *Aphis spiraecola*. Aphid counting was carried out on 12 leaves for each of the six species retained (clementine, lemon, grapefruit and three varieties of mandarin), during autumn (October 2014) and spring (April 2015) flushes. In addition, mineral contents of the leaves in P, K, Na, Ca and Li were measured for the same periods. The results showed that the infestation levels of the studied varieties were higher in the spring flush than in the autumn one. Moreover, analyzes of young leaves showed an important intraspecific (mandarin varieties) and interspecific differences in the mineral composition between the examined citrus trees. The study of the relationship between infestation levels by *A. spiraecola* and mineral content of the six examined species showed no significant correlation, suggesting a marginal role of the five analyzed minerals in the relation citrus – *A. spiraecola*.

Key words: citrus aphid; clementine; lemon; grapefruit; mandarin; flushes

Razmerje med pojavljanjem jabolčne uši *Aphis spiraecola* Patch, 1914 (Hemiptera: Aphididae) in mineralno sestavo listov citrusov

Izvleček: Spomladanski in jesenski viški rasti citrusov so navadno obdobja njihove največje okužbe z listnimi ušmi, vendar je znano zelo malo o pomenu mineralne sestave listov na njihovo pojavljanje. Namen prispevka je bil preučiti korelacijo med nekaterimi minerali v listih različnih citrusov in stopnjo okužbe z listno ušjo *Aphis spiraecola*. Štetje listnih uši je bilo izvedeno na 12 listih vsake od preučevanih vrst (klementine, limone, grenivke in treh sort mandarine), v jesenski (oktober 2014) in spomladanski (april 2015) rasti. Dodatno so bile v istem obdobju v listih izmerjene vsebnosti P, K, Na, Ca in Li. Rezultati so pokazali, da je bila stopnja okužbe pri vseh sadnih vrstah večja v obdobju spomladanske kot jesenske rasti. Analize mladih listov so še pokazale pomembne znotrajvrstne razlike (med sortami mandarin) in medvrstne razlike v mineralni sestavi pregledanih citrusov. Raziskava odvisnosti med velikostjo okužbe z vrsto *A. spiraecola* in mineralno sestavo analiziranih vrst citrusov ni pokazala značilne korelacije, kar kaže na marginalno vlogo petih analiziranih mineralov v razmerju citrusov in preučevane listne uši.

Ključne besede: listne uši citrusov; klementina; limona; grenivka, mandarina; viški rasti

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1 INTRODUCTION

Herbivorous insects identify their host plants through the morphological aspect, chemical signals and sometimes by the combined action of all these factors (Städler & Reifenrath, 2009). Upon herbivore attack, plants produce and emit volatile organic compounds, and some of them may be used in defensive strategy namely the attraction of the herbivores natural enemies (Laznik & Trdan, 2018). In addition, the performance of insects is determined directly by the quality of host plants (Sun & Ge, 2011). The main nutritional needs of insects are amino acids, vitamins, minerals, carbohydrates, lipids and sterols (Silva et al., 2005).

There is much evidence in the literature about the importance of minerals in plant resistance (Hedin et al., 1977). For instance, Khattab (2007) reported that potassium may play a role in the defense mechanism of aphid-infested plants. Several authors have mentioned that low K levels have a positive effect on aphids (Myers et al., 2005; Myers & Gratton, 2006; Hayes et al., 2009), because a lack of potassium in plants favors the accumulation of amino acids in tissues (Amtmann & Armengaud, 2009; Soetan et al., 2010). Mineral ions are important to insect's physiology in at least three major processes: enzyme activation (K, Mg, Fe, Co, Mn), trigger and control mechanisms (Na, Ca, K), and structure formation (Mg) (Silva et al., 2005). Moreover, pests need adequate quantities of several minerals to grow and reproduce (Wigglesworth, 1966). Components such as carbon and nitrogen act directly on the fertility of the pest (Awmack & Leather, 2002).

Shoot growth occurs in most types of citrus in well-defined waves (flushes). The spring flush is the most important one, containing both vegetative and reproductive shoots (Spiegel-Roy & Goldschmidt, 1996). According to Lotmani et al. (2008), the chemical composition of the leaves formed during the different flushes is generally different.

Previous studies (Lebbal & Laamari, 2015; Lebbal & Laamari, 2016) have shown that spring and autumn flushes are the most infested by aphids. Nevertheless, research on the effect of the mineral composition of citrus leaves on aphids is almost absent. Therefore, this paper aims to study the correlation between certain leaf minerals of some citrus varieties and their infestation level by *Aphis spiraecola* Patch, 1914.

2 MATERIALS AND METHODS

In order to show the effect of the leaf chemical composition on the infestation of six citrus species (clementine clone 63, lemon 'Eureka', grapefruit 'Shambar' and three varieties of mandarin: 'Ortanique', 'Carvalhal' and 'Commune'), aphid counting was carried out on 12 randomly chosen young leaves belonging to 4 trees for each of the retained varieties, at the rate of 3 leaves / tree, distributed over the different cardinal directions. These leaves were collected during autumn (October 2014) and spring (April 2015) flushes. Moreover, a foliar analysis of healthy young leaves of these same periods was realized. In total, five minerals were quantified: phosphorus (P) using colorimetry, and sodium (Na), potassium (K), calcium (Ca) and lithium (Li) using flame photometry (Jenway, model PFP7). This latter offers interesting possibilities for the study of the mineral composition of plants (Gueguen & Rombauts, 1961). The location of the young leaves taken was at the periphery of the foliage of the analyzed varieties (Martin-Prével et al., 1965), at about the height of a person. These leaves were dried and then they were crushed for later use in the determination of mineral elements.

The studied orchard (36° 42' N ; 6° 47' E ; 200 m above sea level) is situated in Skikda province (northeast of Algeria) characterized by a sub-humid climate. Its trees were planted in 2001. They were subject to almost the same technical itinerary. The used stock for graft is Troyer citrange (*Citrus sinensis* L. × *Poncirus trifoliata* Raf.) except for lemon which is grafted on volkamer lemon (*Citrus volkameriana* Pasquale). The studied orchard has been managed with limited spraying of pesticides. Weeding was performed mechanically and irrigation was applied during the dry season. Whereas, the fertilization was carried out using 46 % urea.

A correlation analysis was carried out between the level of citrus infestation and mineral content of leaves during the autumn and spring flushes. These analyzes were performed using SPSS software for Windows 10.0.5 (SPSS, Inc.).

3 RESULTS AND DISCUSSION

It was noticed that the infestation levels of these varieties were higher in the spring flush than in the autumn one (Table 1). In addition, analyzes of young leaves taken during the two flushes showed remarkable intraspecific and interspecific differences in the mineral composition. Plants do not have the same mineral requirements. Their contents in these elements affect their physiology and consequently the herbivorous insects feeding on them (Silva et al., 2005). Several authors, among others, Marchal et al. (1974), Roversi et al. (2008) and Pasković et al. (2013), indicated differences in leaf composition in nutrients for different fruit trees.

In general, concentrations of lithium and phosphorus are higher during the spring flush than in the autumn one.

The study of the relationship between degrees of aphid infestation and foliar content of the six examined cultivars showed no significant correlation (Table 2). Similarly, Harrewijn (1970) found that difference in longevity and reproduction rate of *Myzus persicae* (Sulzer, 1776) was not correlated with the total N or soluble N-content of the potato leaves.

Silva et al. (2005) revealed that variation in aphid abundance along different sampling times is correlated to C : N ratio, N, Mg, P and S, but correlations vary with cultivar and aphid species. For instance, they found no significant correlations between aphid population variation and minerals for an alfalfa resistant cultivar, except for C : N ratio. Likewise, Myers et al. (2005) observed no significant difference in mean generation time between soybean aphids feeding on the K-deficient and non-deficient soybean leaves. Nevertheless, they indicated that aphids in the K-deficient treatment exhibited significantly greater intrinsic rate of increase, finite rate of increase, and net reproductive rate relative to aphids feeding on non-deficient leaves. The yellowing associated with potassium deficient soybean leaves may preferentially attract migrating soybean aphids, placing potassium deficient fields at a further disadvantage (Hogg & Gratton, 2010).

Many correlations have been reported between some minerals and biotic parameters of aphids in subsequent studies (Douglas & van Emdeen, 2007; Djazouli, 2010; Agarwala & Das, 2012; Helfenstein et al., 2015). Miyasaka et al. (2007) mentioned that increased reproduction by *Sipha flava* (Forbes, S.A., 1885) aphids on kikuyu (*Pennisetum clandestinum* Hochst) was accompanied by high foliar N. Moreover, short development times of *Macrosiphum euphorbiae* Thomas, 1878 were associated with high P and K content in *Petunia* leaves (Jansson & Ekbom, 2002).

In the present study, aphid colonies may be affected much more by other factors (climate, primary and secondary metabolites) than by the leaf composition in these mineral elements. According to Jansson and Ekbom (2002), the complexity of plant nutrient content on aphid performance suggests that not only nutrient levels but also ratios of nutrients should be considered.

In addition, interactions between nutrients and allochemicals may be key factors in plant susceptibility to

Table 1: Variation in infestation levels (mean number ± standard error of *A. spiraecola* aphids/leaf) and mineral content (in μg g^{-1} of dry matter) of citrus leaves during the autumn and spring flushes

Flush	Parameters	Clementine Clone 63	Lemon Eureka	Grapefruit Shambar	Mandarin Ortanique	Mandarin Carvalhal	Mandarin Commune
Autumn	Infestation	13.75 ± 10.10	5.92 ± 4.91	1.58 ± 0.92	5 ± 1.51	12.83 ± 3.36	8.67 ± 4.23
	Na	0.5	0.58	0.54	0.55	0.56	0.66
	P	3.4	5.7	5.7	4.7	5.2	61.8
	Li	38	38	43.1	32.2	32.2	26.5
	K	29.4	63.4	41.8	56.4	71.8	199.9
	Ca	23071.4	9137.1	18507.9	8785.7	3428.5	1642.8
Spring	Infestation	71.08 ± 14.84	33 ± 18.98	50.17 ± 25.80	48 ± 9.90	20.83 ± 11.45	12.08 ± 5.83
	Na	0.56	0.55	0.57	0.61	0.56	0.57
	P	11.2	13.3	14.1	14.9	5.7	5.5
	Li	43.8	38	49.5	95.6	112.9	107.2
	K	52.4	60.2	62.7	92.9	71.4	73.9
	Ca	15928.5	8785.7	7000	17714.2	17714.2	12357.1

Table 2: Coefficients of correlation between the level of infestation of six citrus varieties by *A. spiraecola* and the mineral contents of their leaves

Analyzed elements Infestation degree	Na	P	Li	K	Ca
Correlation of Pearson	0.042	- 0.011	0.153	- 0.129	0.181
P	0.896	0.974	0.635	0.690	0.573

insect attack (Reese, 1983). Some allochemicals may even make certain nutrients not assimilable (Reese, 1977).

4 CONCLUSION

This study revealed that, compared with the autumn flush, the six citrus species tested had a higher aphid's infestation rate during the spring period. Furthermore, clear differences were shown in mineral composition between the examined varieties. However, statistical analysis showed no significant correlation between aphid infestation levels during these two periods and young leaf content in mineral elements (P, K, Na, Ca and Li). Further studies are desirable in this field, in order to clarify the direct and indirect contribution of each mineral element in the resistance or sensitivity of citrus to aphid attacks.

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