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MORPHOLOGICAL CHARACTERIZATION OF TWO POPULATIONS OF *CIRCULIFER* (HOMOPTERA : CICADELLIDAE) FROM ISRAEL*

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ABSTRACT

General morphology and structure of the male genitalia and female seventh sternum were examined for two wild populations of the genus ***Circulifer.*** The population which was identified as ***Circulifer opacipennis*** complex inhabits prickly saltwort (***Salsola kali***) on the sandy soil of the Mediterranean coast and the other, which was identified as ***Circulifer haemaloceps*** complex, inhabits common goosefoot (***Chenopodium album)*** in the Golan Heights. The two populations did not differ in colour. The internal concealed male genitalia in the population collected from ***S. kali*** were significantly larger than those in the population from **C.** ***album.*** Styles of both populations clearly differed in shape and angle of their apices. The two populations differed also in the size of female 7th abdominal sterna.

KEY WORDS: Classification, *Circulifer* ***spp.,*** *Neoaliturus* ***spp.,*** *Circulifer haematoceps* ***complex,*** *Circulifer opacipennis* ***complex,*** *Salsola kali, Chenopodium album.*

INTRODUCTION

Identification of Cicadellidae is mostly based on differences in the structure of the male genitalia. However, for certain leafhopper groups, identification based on the above is insufficient. A typical case occurs in the genera *Circulifer* Zakhvatkin and *Neoaliturus* Distant (Oman, 1970). Linnavuori (1962) reported on the identification of *Neoaliturus haematoceps* ssp. *opacipennis* from Israel. He apparently referred to either *C. haematoceps* (Mulsant and Rey) or *C. opacipennis* (Lethierry), which were reported from the Mediterranean by Young and Frazier (1954). Oman (1970) recommended that the genus *Neoaliturus* should be separate from *Circulifer,* based on several characters including the extent of the closure of the circle formed by the rami of the aedeagus: *Circulifer* with complete circles and *Neoaliturus* with incomplete circles. Swirski (1959) identified *Circulifer fenestratus* (Herrich-Shaffer) from several areas in Israel. This species, according to Oman (1970), belongs to *Neoaliturus* and not *Circulifer.* Several morphologically distinct groups in the *Circulifer tenellus* (Baker) complex were discerned in Israel (Klein and Almeida, 1990; Klein and Raccah, 1987; Klein et al., 1982). Young and Frazier (1954) found several species of *Circulifer* from the Mediterranean when analysing collections made during a survey in several Mediterranean countries (Frazier, 1953). They identified *C. tenellus* and *C. dubiosus* (Matsumura) and noted that the latter was similar to C. *tenellus* in many morphological aspects. These two species are characterized by oblong or oval male genital plates (Young and Frazier, 1954). They also identified two common Old World species, C. *opacipennis* and *C. haematoceps,* which have triangular male genital plates (Young and Frazier, 1954). Two less

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common species, *C. nausharensis* (Pruthi) from North India and *C. nitidus* (Young and Frazier) from Spain, both closely related to *C. opacipennis*, were also reported in the study.

Recent morphological reassessment of 41 populations of old and fresh *Circulifer* specimens from different parts of the world, made by M. W. Nielson (personal communication), indicated 4 major species complexes. Species within each complex could not be identified with certainty.

Circulifer and *Neoaliturus* species are known as vectors of plant pathogens in the USA (Oldfield et al., 1976, 1977; Severin, 1921), in the Near East (Bennet and Tanrisever, 1957; Kheiry and Alimoradi, 1969) and in the Mediterranean area (Bove et al., 1988; Fos et al., 1986; Klein, 1970; Klein et al., 1988). In the course of searching for the vectors of disease pathogens in Israel, two leafhopper populations characterized by triangular plates in the males were found to occur in large numbers. One was collected on prickly saltwort (*Salsola kali* L.) and the other on common goosefoot (*Chenopodium album* L.). Their host preferences have recently been investigated in the laboratory (Klein and Raccah, 1991). This paper attempts to separate these two leafhopper populations on the basis of the male genitalia and female 7th abdominal sterna.

MATERIALS AND METHODS

Surveys for *Circulifer* species were carried out in different regions of Israel in order to assess their distribution. Samples were brought to the laboratory to study their role as vectors of plant pathogens. Two populations which were found in large numbers and seemed to be distinct from each other required further examination. One population was found on *S. kali* in Rishon LeZiyyon and the secomd was found on *C. album* in the Golan Heights.

Abdomens of twelve males and six females from each of the two populations were cleared in 10% KOH solution. The male genital structures were separated and mounted on microscope slides, for further illustration and measurements. In the females, the 7th abdominal sternum was removed and processed similarly. These structures were examined using a compound microscope at ×250 and measured for a comparative study. A schematic diagram of the male aedeagus, connective, style, plate and the female seventh sternum is given in Fig. 1 (B, C, D, E and A, respectively). The lower case letters and lines represent areas where measurements were most readily taken for these comparison data. The mean (±SD) length of various measurements of the genitalia are given in microns in Table 1. Data were analyzed statistically by the Student's *t*-test.

RESULTS

Comparisons of the two *Circulifer* populations were made on external morphology, male genitalia and the female 7th sternum.

Circulifer population from *S. kali* (*C. opacipennis* complex): Dorsal side is mostly yellowish green with no special marks. Forewings hyaline with a yellowish tinge, veins somewhat darker yellow becoming brown near the apical junction with marginal veins.

Length of males 3.7–4.1 mm, of females 4.1–4.2 mm (n = 20). Male with genital plates triangular (Fig. 2D). Aedeagus with rami usually forming a full circle (Fig. 2B); sometimes circle is opened during preparation. In lateral view, in all cases examined, the apex of the rami crossed the lateral margins of the aedeagus. Styles have their apices sharply angled laterally (Fig. 2C).

The posterior margin of the female 7th abdominal sternum has a large medial emargination. The lateral edges of the emargination are further indentated, and the lateral posterior margins protrude further than the lateral margins of the sternum (Fig. 2A).

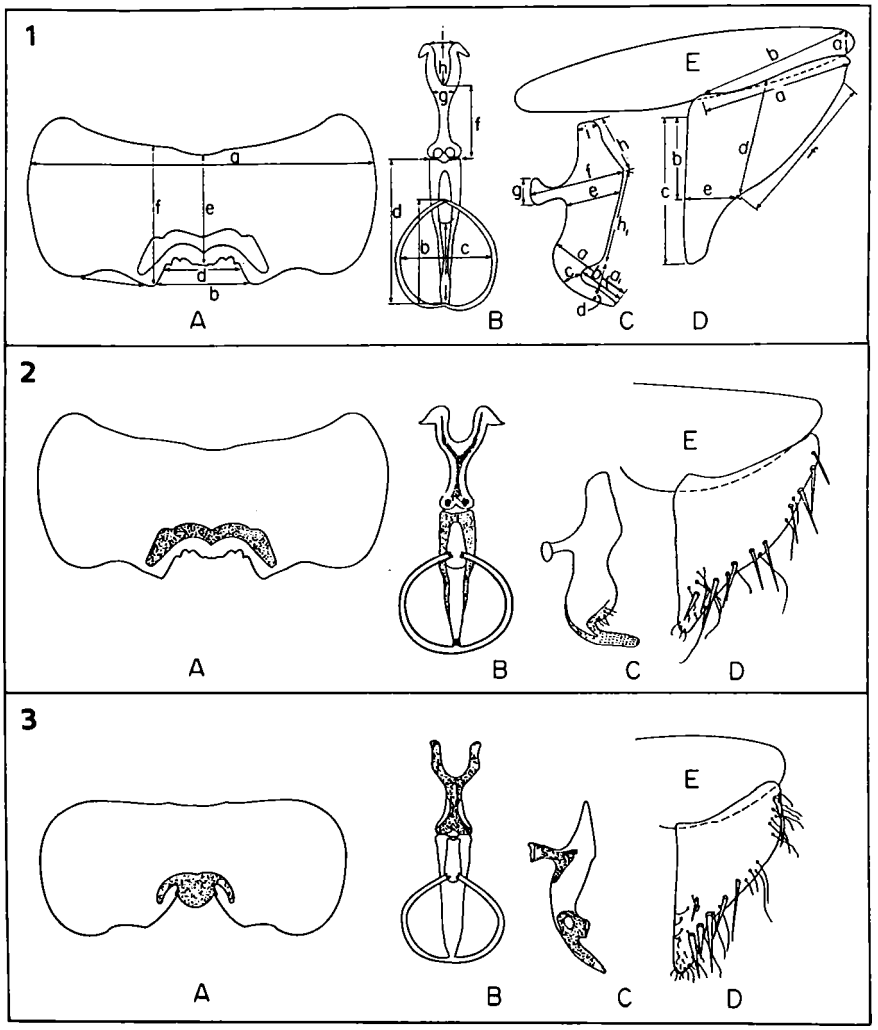
Circulifer sp. from *C. album* (*C. haematoceps* complex): Dorsal side yellowish green, unmarked. Forewing hyaline yellowish green, being darker than the background and gray in color close to the wing's external margin.

Length of males 3.5–4.0 mm, of females 3.8–4.0 mm (n = 20). Male genital plates triangular

TABLE 1
Differences in mean lengths (μm ± SD) of distinct measurements of the female seventh sternum (A) and male genitalia (B to E) of two *Circulifer* populations (6 females, 12 males) from Israel

	Females			Males										
	Seventh sternum (A)			Aedeagus and connective (B)			Style (C)			Genital plate (D)			Valve (E)		
										Leafhoppers trapped on					
	S. kali	C. album		*S. kali*	*C. album*		*S. kali*	*C. album*		*S. kali*	*C. album*		*S. kali*	*C. album*	
a	636.9(30.20)	588.8(59)	NS	145.4(14.3)	106.0(20.0)	VS	136.7(12.3)	146.9(14.3)	NS	262.7(14.8)	208.4(11.1)	VS	56.8(10.2)	64.5(8.2)	NS
a_I							88.6(2.5)	91.1(6.7)	NS						
b	187.4(12.8)	160.0(11.3)	S	211.5(9.2)	192.0(22.5)	S	28.7(2.6)	16.9(3.6)	VS	181.8(22.0)	154.6(13.8)	VS	291.3(20.5)	261.12(10.2)	VS
c	180.2(13.8)	145.9(15.4)	S	250.0(17.4)	198.7(30.7)	VS	52.2(4.1)	34.8(4.1)	VS	294.9(17.9)	262.7(16.9)	VS			
d	86.0(11.8)	55.0(7.7)	VS	274.9(15.3)	257.0(13.8)	S.	25.6(0)	26.6(4.1)	NS	158.2(7.7)	160.3(22.0)	NS			
e	214.0(21.0	230.4(32.3)	NS	67.6(4.6)	57.8(3.4)	VS	76.8(4.6)	64.5(3.6)	VS	171.0(11.3)	93.7(6.8)	VS			
f				122.4(5.6)	99.3(12.3)	VS	161.8(10.2)	134.7(4.9	VS	284.7(21.0)	231.9(17.0)	VS			
g				81.4(8.5)	73.2(8.2)	S	43.0(6.1)	41.5(5.1)	NS						
h				76.3(7.2)	69.1(6.9)	NS	102.4(8.7)	99.8(15.9)	NS						
h_I							134.7(14.8)	119.3(15.9)	NS						
i				98.8(12.3)	86.5(16.4)	S	41.5(4.6)	30.2(4.6)	VS						

Statistical differences: NS = Not significant at $P > 0.05$. S = Significant at $P < 0.05$. VS = Very significant at $P < 0.01$.



Figs. 1–3. *Circulifer* spp. 1. General scheme. Structure designated A in Fig. 1–3 represents the female 7th abdominal sternum. Structure designated B, C, D and E in Fig. 1–3 belongs to the male genitalia (B = aedeagus and connective, C = style, D = genital plate and E = valve). 2. Male and female structures of the *Circulifer* (*C. opacipennis* complex) population collected on *Salsola kali*. 3. Male and female structures of the *Circulifer* (*C. haematoceps* complex) population collected on *Chenopodium album*.

(Fig. 3D). Aedeagus with rami forming a full circle (Fig. 3B). Styles with their apices slightly bent (Fig. 3C). The posterior margin of the female 7th abdominal sternum with a narrow median emargination. The lateral edges of the emargination sharply indentated, and the lateral posterior margins protruding less than the lateral margins of the sternum (Fig. 3A).

Female seventh sternum: Measurements of 'd' (Fig. 1A) in the emargination for the specimens from *S. kali* (*C. opacipennis* complex), given in Fig. 2A are much longer ($P < 0.01$) than those for specimens from *C. album* (*C. haematoceps* complex) (Table 1 and Fig. 3A). The measurements

of 'b' and 'c' are also larger in specimens from *S. kali* (Fig. 2A) than from *C. album* ($P < 0.05$) (Fig. 3A; Table 1). No significant difference ($P > 0.05$) was found between the two population comparing measurement 'a' and 'e'.

Male genitalia: The measurements of the aedeagus marked 'a', 'c', 'e' and 'f' (Fig. 1B) are much longer in specimens from *S. kali* (Fig. 2B) than in specimens from *C. album* (Fig. 3B) (Table 1). The measurements of the aedeagus and connective (Fig. 1B, 'b', 'd', 'g' and 'i') are also longer in specimens from *S. kali* than in specimens from *C. album* (Table 1). The measurements of the style designated 'b', 'e', 'f' and 'i' (Fig. 1C) are much longer in specimens from *S. kali* (Fig. 2C) than in specimens from *C. album* (Fig. 3C) (Table 1). Other measurements do not differ significantly in either population. The measurements in the plates designated 'a', 'b', 'c', 'e' and 'f' (Fig. 1D) are much longer in specimens from *S. kali* (Fig. 2D) than from *C. album* (Fig. 3D) (Table 1). The length of the valve (Fig. 1E) from the basal lateral margin to its posterior center ('b') is significantly longer in specimens from *S. kali* (Fig. 2E) than in those from *C. album* (Fig. 3E) while the width of the internal margin ('a') is about the same in the two leafhopper populations (Table 1).

DISCUSSION

There is a great need to improve the taxonomy of *Circulifer* spp. The wide distribution and vectorial capacity of *Circulifer* species demands a more reliable way to identify species. The beet leafhopper, *C. tenellus*, considered the most destructive leafhopper in western USA since the beginning of this century, is a very efficient vector of the curly top virus, a disease which causes large yearly losses in sugar beet, tomato, beans, cantaloupe and other crops (Severin, 1921). The beet leafhopper is also the natural vector of *Spiroplasma citri*, the causal agent of the 'stubborn' disease of citrus and of the beet leafhopper-transmitted virescence agent (BLTVA) in the USA (Oldfield et al., 1976, 1977).

Diseases with an etiology and agents similar to the American beet curly top virus (BCTV) and the citrus 'stubborn' disease were also found in the Mediterranean area and the Near East. BCTV has been observed in sugar beets in Turkey and Iran and was transmitted experimentally by *C. haematoceps* (Bennett and Tanrisever, 1957; Kheyri and Alimoradi, 1969). On the other hand, stubborn disease is widespread along the coast of eastern Mediterranean and North African countries. Fos et al. (1986) successfully transmited the Moroccan isolate of *S. citri* by a leafhopper identified as *C. haematoceps*.

The American *C. tenellus* is the only known species of the *Circulifer* in the USA (Oman, 1970). In the Old World, *Circulifer* and *Neoaliturus* are represented by numerous species which so far have not been adequately separated from each other by conventional methods.

Young and Frazier (1954) revised the genus *Circulifer*. In their study, the male plates in *C. tenellus* and *C. dubiosus* were similar but differed from those in *C. haematoceps*. Their studies on *C. tenellus* and *C. dubiosus* required description of subspecies (*C. tenellus tenellus* and *C. tenellus ambiguosus*; *C. dubiosus dubiosus* and *C. dubiosus infirmus*) in order to separate the various populations. However, two species, namely *C. haematoceps* and *C. opacipennis*, were not subdivided into subspecies.

The present morphological study has shown significant differences in length of different measurements of the structures of the male genitalia and female 7th abdominal sclerits in two populations of *Circulifer*. Recent studies showed that these two leafhopper populations had distinct survival rates on different host plants (Klein and Raccah, 1991). The populations which were characterized in this study are believed to be complexes which explain our difficulty in assigning them to a particular species name. The taxonomic status in this genus led M.W. Nielson (personal communication) to suggest the use of the term "complex" for at least 4 major groups: *tenellus*, *dubiosus*, *haematoceps* and *opacipennis*. In the present paper, we followed his line of

thought and therefore have placed the two populations in two different species complexes, namely the *C. haematoceps* complex and the *C. opacipennis* complex.

Correct identification of *Circulifer* vector species is extremely important in studies of the stubborn disease and the curly top disease agents in the Near East. *Circulifer haematoceps* identified in transmission studies by Fos et al. (1986) was collected from *S. kali* and from *Matthiola incana* in Syria. In Israel, two *Circulifer* populations were trapped in large numbers. One was collected from *S. kali*; it was found to be monophagous on this host and failed to breed on any other plant species tested in the laboratory (Klein and Raccah, 1991). Young and Frazier (1954) indicated that the *C. haematoceps* which was collected in the Mediterranean survived only on *Cistus* plants. On the other hand, the *C. haematoceps* from Syria was collected from several different host plants (Fos et al., 1986). The population which we collected on *S. kali* in Israel was identified by M.W. Nielson (personal communication) as *C. opacipennis* complex; it has shown a greater similarity to the original *C. opacipennis* than to *C. haematoceps* reported by Young and Frazier (1954). Finally, the second population, collected on *C. album*, has a wider host range than the population of *C. haematoceps* from *S. kali* (Klein and Raccah, 1991). This population appears to match the description of the population of *C. haematoceps* reported by Young and Frazier (1954).

We conclude from this and earlier studies of *Circulifer* species that imprecise identification could lead to an improper determination of the vector species and its ecological niche. A comprehensive taxonomic revision of the fauna of *Circulifer* and *Neoaliturus* of the Mediterranean basin and adjacent areas is urgent. It seems that progress will be achieved only in the future, when more sophisticated techniques, such as restriction fragment polymorphism (RFLP), isozymes, and courtship-sound signals are added to the conventional taxonomy of these species.

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REFERENCES

Ball, E. 1907. The genus *Eutettix* with special reference to *E. tenella*, the beet leafhopper: A taxonomic, biologic and economic study of the North American forms. *Proceedings of Davenport Academic Sciences* 12:27–94.

Bennett, C.W. and Tanrisever, A. 1957. Sugarbeet curly top disease in Turkey. *Plant Disease Reporter* 41:721–725.

Bove, J.M., Fos, A., Lallemand, J., Raie, A., Saillard, C. and Vignault, J.C. 1988. Epidemiology of *Spiroplasma citri* in the Old World. *Abstract of 10th International Organization Citrus Virologists Conference, Riverside, California.* pp. 1–98.

Fos, A., Bove, J.M., Lallemand, J., Saillard, C., Vignault, J.C., Ali, Y., Brun, P. and Vogel, R. 1986. La cicadelle *Neoaliturus haematoceps* Mulsant and Rey est vecteur de *Spiroplasma citri* en Méditerranée. *Annalles de Microbiologie* 137:97–107.

Frazier, N. W. 1953. A survey of Mediterranean for the beet leafhopper. *Journal of Economic Entomology* 46:551–554.

Kheyri, M. and Alimoradi, I. 1969. The leafhoppers in sugarbeet in Iran and their role in curly top virus disease. Sugar Seed Institute, College of Agriculture, Karaj Entomology Research Division, Teheran.

Klein, M. 1970. Safflower phyllody — a mycoplasma disease of *Carthamus tinctorius* in Israel. *Plant Disease Reporter* 54:735–738.

Klein, M. and Almeida, L. 1990. Two members of the *Circulifer tenellus* complex exhibit differences in vectoring *Spiroplasma citri*. *Abstracts of the Third International Workshop on Leafhoppers and Planthoppers of Economic Importance. August 13–17, 1990, Wooster, OHIO, USA.*

Klein, M. and Raccah, B. 1987. Morphological variation in *Neoaliturus* spp. (Cicadellidae) from various host plants in Israel. In *Proceeding of the Second International Workshop on Leafhoppers and Planthoppers of Economic Importance.* (July, 28–August 1, 1986, Provo, UTAH, USA). Edit. M.R. Wilson and L.E. Nault. Commonwealth Institute of Entomology, London. p. 267.

Klein, M. and Raccah, B. 1991. Separation of two leafhopper populations of the *Circulifer haematoceps* complex on different host plants in Israel. *Phytoparasitica* 19:153–155.

Klein, M., Raccah, B. and Oman, P.W. 1982. The occurrence of a member of the *Circulifer tenellus* species complex (Homoptera : Cicadellidae : Euscillini) in Israel. *Phytoparasitica* 10:237–240.

Klein, M., Rasooly, R. and Raccah, B. 1988. Transmission of *Spiroplasma citri*, the agent of citrus stubborn, by a leafhopper of the *Circulifer tenellus* complex in Israel. *Proceedings International Citrus Congress, Middle East, Tel Aviv* 2:49.

Linnavuori, R. 1962. Hemiptera of Israel. III. *Annalles Zoologici Societatis Fennicae 'Vanamo'* 24:1–108.

Oldfield, G.N., Kaloostian, G.H., Pierce, H.D., Calavan, E.C., Granett, A.L. and Blue, R.L. 1976. Beet leafhopper transmits citrus stubborn disease. *California Agriculture* 30(6):15.

Oldfield, G.N., Kaloostian, G.H., Pierce, H.D., Granett, A.L. and Calavan, E.E. 1977. Beet leafhopper transmits virescence of periwinkle. *California Agriculture* 31(6):14–45.

Oman, P. 1970. Taxonomy and nomenclature of the beet leafhopper, *Circulifer tenellus* (Homoptera: Cicadellidae). *Annals of the Entomological Society of America* 63:507–512.

Severin, H.H.P. 1921. Minimum incubation periods of causative agent of curly top in beet leafhopper and sugar beet. *Phytopathology* 11:424–429.

Swirski, E. 1959. Contribution to the phenology of leafhoppers (Homoptera) in Israel. Ministry of Agriculture, Agricultural Research Station, Institute of Plant Protection, Department of Horticultural Entomology, Bet Dagan, Israel (in Hebrew).

Young, D.A. and Frazier, N.W. 1954. A study of the leafhopper genus *Circulifer* Zakhvatkin (Homoptera: Cicadellidae). *Hilgardia* 23:25–52.