JARQ 47 (4), 365 - 369 (2013) http://www.jircas.affrc.go.jp

REVIEW

Ecology of the Maize Orange Leafhopper, *Cicadulina bipunctata* (Melichar) (Hemiptera: Cicadellidae)

Keiichiro MATSUKURA* and Masaya MATSUMURA

Agro-Environment Research Division, NARO Kyushu Okinawa Agricultural Research Center (Koshi, Kumamoto 861-1192, Japan)

Abstract

Recent global warming has caused the maize orange leafhopper, *Cicadulina bipunctata* (Melichar), to proliferate in Kyushu, Japan. This leafhopper feeds on several species of poaceous plants. Some host plants such as maize, rice, wheat and oats show abnormal growth, characterized by stunted growth and swelling (upheaving) of leaf veins when attacked by *C. bipunctata*. This abnormal growth is not caused by a virus or phytoplasma, but by the injection of chemical(s) from the leafhopper into the plants. In central and southern areas of Kyushu, serious damage arising from this abnormal growth is known as maize wallaby ear symptom (MWES) and occurs on forage maize during the second cropping season (from late June to November). The degree of MWES is highly dependent on the level of infestation (period and density) by *C. bipunctata*, and since *C. bipunctata* density is quite low until mid-July, the earlier seeding of the second crop and the use of MWES-resistant varieties is an effective way to avoid damage from MWES. This leafhopper shows a unique relationship between plant and herbivores because MWES induction by adult *C. bipunctata* helps improve the development of their offspring through the accumulation of amino acids on the host plant.

Discipline: Insect pest
Additional key words: forage crop, gall, global warming, insect pest, maize wallaby ear symptom

Introduction

Since the 1980s, stunted growth associated with maize wallaby ear symptom (MWES) has been observed in maize, *Zea mays* (L.) in East Asian countries[1,7,10,20]. This symptom is characterized by stunted growth together with severe swelling of leaf veins[12,22] and has threatened maize production in these areas due to substantially reduced yields. Because MWES superficially resembles symptoms caused by viral pathogens, it was initially identified as a viral disease transmitted by the maize orange leafhopper, *Cicadulina bipunctata* (Melichar)[1]. More recent experiments indicated that MWES is caused not by a virus but by chemical(s) injected by *C. bipunctata* during feeding activity[12,22].

In Japan, MWES has been a serious concern since 2000 in Kyushu island, the most southern of the four main islands of Japan. Here, most farmers crop forage maize twice yearly (first crop: April to mid-July; second crop: late July to November), and MWES occurs in the second crop[15].

In this review, we describe the ecology of *C. bipunctata* in temperate Japan and related works from agricultural and scientific perspectives.

Recent outbreak of *C. bipunctata*

This leafhopper was originally distributed in tropical and subtropical regions of the Old World[25]. Kumamoto Prefecture in central Kyushu was the northern distribution limit of *C. bipunctata* until the early 1980s[2,21]. After the late 1980s, this leafhopper was found in all seven prefectures of Kyushu island (Fig. 1)[6,11,23] and is now distributed in most lowlands of Kyushu except the northern part.

The recent rapid expansion of the distribution of *C. bipunctata* is thought to be caused by global warming[17,20]. We examined the effects of climatic factors (temperature, precipitation and sunlight) on the occurrence of *C. bipunctata*, using field data from 2004 to 2010 in Kumamoto Prefecture[17]. The results of a multiple linear regression analysis indicated the increased occurrence of adult *C. bipunctata* due to higher temperatures during both the previous winter and early summer of the current year. Furthermore, unlike most insects distributed in temperate regions, this leafhopper showed no arrested development,

*Corresponding author: e-mail mtkr@affrc.go.jp
Received 8 November 2012; accepted 1 February 2013.

Fig. 1. Current distribution of *C. bipunctata* (shown with shaded areas) in Kyushu island, Japan
Data was based on previous studies[6,11,23].

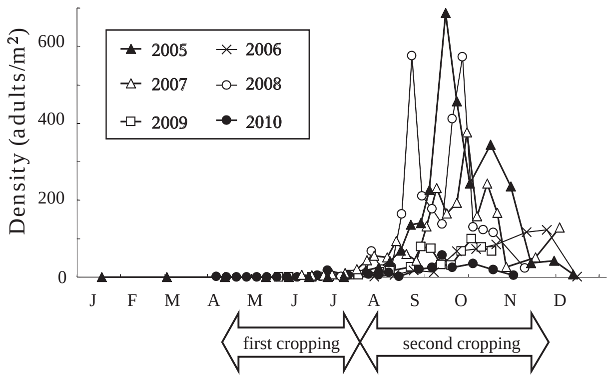

Fig. 2. Seasonal occurrence of *C. bipunctata* on poaceous weeds in Kyushu (Kikuchi, Kumamoto)
Numerous *C. bipunctata* emerge during the second cropping season while few *C. bipunctata* occur in the first cropping.
Data was partially referred to from the previous study[11].

even at 30°C[24]. These data strongly suggest that the recent increased temperatures (average winter temperature in Kyushu has increased approximately 2°C within the past forty years) caused the recent outbreak of *C. bipunctata* in temperate Japan.

Seasonal occurrence

The occurrence of *C. bipunctata* in Kumamoto, Japan fluctuates among seasons (Fig. 2). Meanwhile, the adult density is quite low from winter to early summer (early July), increases from late July, and peaks in September or October. No nymphs were observed during the overwintering season (from January to April) whereas overwintering adults were often collected[11], indicating that this insect overwinters at the adult stage in Japan. The quick increase in *C. bipunctata* density is also observed in temperate China (Sichuan Province)[7].

There is a higher incidence of MWES in the second crop relative to the first due to the higher occurrence of the pest in summer rather than spring (Fig. 2). The density of *C. bipunctata* remains quite low from April to early July, during the first crop, but increases during the seeding period of the second crop (from late July to mid-August) and peaks in September or October, when forage maize is in the vegetative and flowering stages. Forage maize in the second cropping season is attacked by much larger numbers of *C. bipunctata*, thereby causing more serious damage from MWES relative to the first crop.

Highly variable occurrence among years is another remarkable characteristic of this insect pest. For example, adult density on poaceous weeds in forage maize fields reached more than 500 adults/m^2 in 2005 and 2008 as opposed to approximately 100 adults/m^2 or fewer in 2006 and 2010[11] (Fig. 2). Similarly, the density of nymphs on poaceous weeds fluctuates substantially from approximately 300 to more than 3,000 nymphs/m^2 from year to year[11]. This yearly fluctuation is mostly temperature-related and would affect the degree of MWES injury in fields.

Host plants

Various poaceous plants are reported as hosts of *C. bipunctata* (Table 1), including several important cereal crops such as barley, *Hordeum vulgare* (L.), foxtail millet, *Setaria italica* (P. Beauv.), maize, rice, *Oryza sative* (L.), Sorghum, *Sorghum bicolor* (Moench.), and wheat, *Triticum aestivum* (L.)[8,10,13]. Foxtail grass, *Setaria viridis* (L.), goose grass, *Eleusine indica* (L.), guinea grass, *Panicum maximum* (Jacq.), italian ryegrass, *Lolium multiflorum* (Lam.), Oats, *Avena sativa* (L.), orchard grass, *Dactylis glomerata* (L.), rhodes grass, *Chloris gayana* (Kunth.), southern crab grass, *Digitaria ciliaris* (Retz.) and sudan grass, *Sorghum sudanense* (Piper.) are also reportedly host plants for this insect[8,13,19], whereas it cannot feed on johnson grass, *Sorghum halepense* (L.) and red sprangletop, *Leptochloa panicea* (Retz)[8,13]. Nymphal survival and developmental

Table 1. Availability of poaceous plants as hosts for C. *bipunctata*

Plant	Survival[a]	Development[b]	Swelling of leaf veins[c]	References
Maize	+	+	+	8, 12
Rice	±	+	+	8, 10
Wheat	+	+	+	4, 8
Oats	+	+	+	13
Millet	+	+	n.d.	8
Sorghum	+	+	–	13
Barley	+	+	–	5, 13
Orchard grass	+	±	–	13
Sudan grass	+	+	–	13
Rhodes grass	±	n.d.	–	13
Guinea grass	±	n.d.	–	13
Italian ryegrass	+	n.d.	+	19
Goose grass	+	n.d.	n.d.	11
Southern crab grass	+	n.d.	n.d.	11
Foxtail grass	+	n.d.	n.d.	11
Bamboo grass	+	n.d.	n.d.	11
Red sprangletop	–	–	–	8
Johnson grass	–	–	–	11

+: positive, ±: positive but not preferable, –: negative, n.d.: no data.
a: *C. bipunctata* fed on or not
b: *C. bipunctata* developed from nymph to adult or not
c: Swelling of leaf veins appeared or not when it was infested by *C. bipunctata*

rates are relatively high on sorghum, sudan grass and oats[8,13], indicating that these plants are preferable hosts for *C. bipunctata*. Conversely, the survival rates of nymphs are relatively low on rice, rhodes grass and guinea grass[8,13], indicating that these plants seem inferior to other host plants. In Kyushu, the main host plants of *C. bipunctata* in forage crop fields and surrounding vegetation differ among seasons. Overwintering adults were collected at most from bamboo grass, *Sasa* spp.[11], although there is no evidence that *C. bipunctata* can multiply on the plant. Some overwintering adults were also found in winter crops (from December to May) of barley, wheat, oats and italian ryegrass[11,19]. On these winter crops, first-generation nymphs were found in May[11,19], suggesting that these crops represent major host plants from overwintering to spring. From late spring to autumn, both adults and nymphs were collected from maize (both the first and second crops) and poaceous weeds such as foxtail grass, goose grass and southern crab grass[11,16]. Numerous *C. bipunctata* were also collected from autumn crops of oats and barley[19] (from September to December).

Feeding by *C. bipunctata* causes stunted growth together with upheaval of leaf veins; not only on maize but also some other poaceous plants (Table 1). When oats were attacked by *C. bipunctata*, the stems do not elongate appropriately and the leaf veins swell[13]. Similar symptoms were also observed on rice, wheat and italian ryegrass[4,10,19]. Despite the stunted growth caused by *C. bipunctata* on these poaceous crops however, the damage is less serious compared to maize, likely because most poaceous crops have a tillering stage, unlike forage maize. Compensation by healthy tillers, which are not attacked by *C. bipunctata*, may mitigate the decline in yield caused by this symptom. No upheaval of leaf veins was observed on barley, sorghum, orchard grass, rhodes grass, sudan grass and guinea grass, although growth stunting was observed to some extent after *C. bipunctata* feeding[5,13,19].

Mechanism of MWES induction

As mentioned above, MWES is caused by chemical(s) from *C. bipunctata*[22]. Upheaval of leaf veins appears not on leaves that are infested by *C. bipunctata* but on those that emerge during or after infestation[12]. The symptom does not appear on any leaves that emerge after *C. bipunctata* are removed from the plant, supporting the previous finding that MWES is not caused by a virus (if it were, the symptom would appear on all leaves emerging after infestation by *C. bipunctata*). These findings suggest that chemical(s) injected from *C. bipunctata* into the leaf act(s) on the shoot apical meristem and/or leaf primordia of the plant and disrupt(s) the morphogenesis of new leaves.

Both the infestation density and period of *C. bipunctata* positively affect the degree of MWES[12]. Both increased infestation density and period resulted in more severe MWES (i.e. greater upheaval of leaf veins and more severe stunted growth). For example, the degree of symptoms was slight after 1-hour feeding by four pairs of adult *C. bipunctata*, but intensified with increasing feeding duration, peaking at 48 hours of feeding[12], which means the MWES induction by *C. bipunctata* is a dose-dependent reaction. Adult males, females and nymphs of *C. bipunctata* can all induce MWES at a similar level[12,14].

The plant stage during the attack by *C. bipunctata* also affects the degree of MWES appearance. When maize is infested by *C. bipunctata* at a younger stage, MWES tends to be more severe. For example, when ten adult male *C. bipunctata* were fed on maize seedlings at the second or third leaf stage, the plant suffered obvious stunted growth with severe swelling of leaf veins. However, when maize past the sixth leaf stage was exposed to the same condition, it scarcely exhibited any MWES[15].

The two characteristics of MWES (i.e. stunted growth and upheaval of leaf veins) seem attributable to different physiological mechanisms. Kumashiro et al.[5] examined the relationship between the degrees of growth stunting and upheaval of leaf veins, using six barley chromosome disomic addition lines of wheat. The two symptoms differed significantly among lines, but no significant correlation between the degrees of growth stunting and upheaval was detected.

Practical control to MWES

Forage crops are cheaper than those for human food, meaning farmers must control MWES economically. There are two effective cultural control methods that incur no extra cost compared to standard cultivation. The first involves cropping MWES-resistant varieties, two of which have been established in Japan. These varieties rarely exhibit MWES, even if attacked by *C. bipunctata*[15]. The occurrence of MWES in fields seemed to decrease after widespread use of resistant varieties, indicating that the use of the latter usually suffices to control MWES. Unfortunately, even resistant varieties exhibit MWES when they are exposed to a major infestation by *C. bipunctata* at the seedling stage[15]. Further improvement of MWES-resistant varieties is required.

Another effective cultural method in Kyushu is early seeding. As described above, the second crop begins from late July to mid-August, when the *C. bipunctata* density begins to increase (Fig. 2). This means that young maize seedlings are attacked by numerous *C. bipunctata*. Because maize larger than the sixth leaf stage is not affected by *C. bipunctata* infestation[15], earlier seeding enhances plant growth under low-density conditions of *C. bipunctata*, and

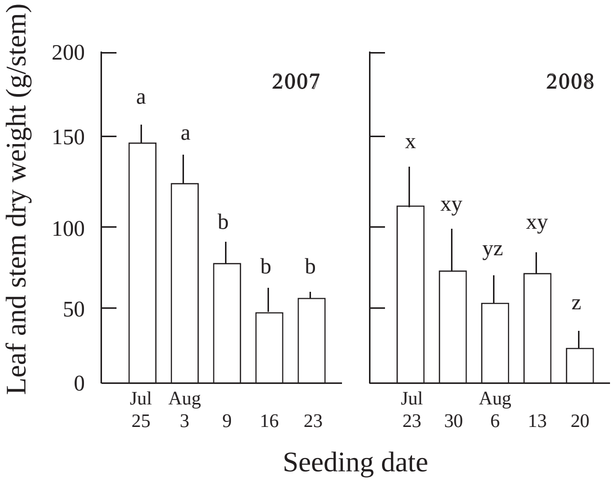

Fig. 3. Effect of seeding date of the second crop on leaf and stem dry weight (Kikuchi, Kumamoto)
a-b, x-z: the same letters above vertical bars, which indicate standard deviation of yield, mean no significant difference in yield among the seeding date the same year (Tukey HSD test, α=0.05).
Data was based on the previous study[15].

decreases the extent of MWES damage. In our field study, the yields of leaves and stem dry matter peaked in maize seeded in late July, and gradually decreased as the seeding date progressed from early to mid-August (Fig. 3). Using early seeding, farmers can cultivate MWES-susceptible varieties in fields where *C. bipunctata* is distributed.

Adaptive significance of the MWES induction for *C. bipunctata*

The induction of MWES on host plants is adaptive to the reproduction of *C. bipunctata*. Nymphs feeding on maize suffering from MWES have higher survival and developmental rates than those feeding on healthy maize[18]. Moreover, the densities of free amino acids (alanine, asparagine and serine, in particular) on leaf blades with leaf vein upheaval significantly exceed those of healthy leaf blades. Because *C. bipunctata* is a phloem-sap feeder (K. Matsukura, unpublished), the higher amino acid content in host plants boosts the development of nymphs[3,9,26]. We suggest that nymphs from host plants with MWES elicited by their parents have higher fitness. This is a unique phenomenon whereby adults help enhance the performance of their offspring indirectly through host plant manipulation.

Conclusion and future direction

Recent global warming has triggered outbreaks of *C. bipunctata* and MWES in East Asian countries. Further global warming may cause both the population density and the distribution area of *C. bipunctata* to expand. Although economic damage has solely been reported in forage maize at present, if the *C. bipunctata* density continues to increase, MWES injury may also affect other important crops such as rice and wheat. Fluctuations in the seasonal occurrence and distribution of this insect pest should thus be monitored carefully.

Host plant manipulation by insects using toxins (chemicals) is another interesting example of insect-plant interaction. Unfortunately, the physiological aspects of MWES induction remain poorly understood. Further analyses using biochemical and molecular biological approaches are expected.

Acknowledgements

Our study was supported by the development of mitigation and adaptation techniques to global warming in sectors of agriculture, forestry, and fisheries (44130) from the Ministry of Agriculture, Forestry, and Fisheries of Japan.

References

1. Grylls, N. E. (1975) Leafhopper transmission of a virus causing maize wallaby ear disease. *Ann. Appl. Biol.*, **79**, 283-296
2. Hayashi, M. (2002) Homoptera. *In* Check List of the Insect of the Ryukyu islands, Flora and Fauna in Okinawa No. 1. 2nd ed, ed. Azuma, S., Biological Society of Okinawa, Okinawa, Japan, 97-112 [In Japanese].
3. Karley, A.J. (2002) Amino acid composition and nutritional quality of potato leaf phloem sap for aphids. *J. Exp. Biol.*, **205**, 3009-3018.
4. Kawano, S. (1994) Occurrence of maize leaf gall disorder induced by the feeding of the leafhopper, *Cicadulina bipunctella* (Matsumura) (Homoptera: Cicadellidae) in Okinawa, Japan. *Okinawaken Nogyoshikenjo Kenkyu Hokoku* (*Bull. Okinawa Agric. Exp. Sta.*), **15**, 51-57 [In Japanese].
5. Kumashiro, S. et al. (2011) Effect of barley chromosome addition on the susceptibility of wheat to feeding by a gall-inducing leafhopper. *Naturwissenschaften*, **98**, 983-987.
6. Kumashiro, S. et al. (2012) Distribution of *Cicadulina bipunctata* (Hemiptera: Cicadellidae) in northern and eastern parts of Kyushu, Japan. *Kyushu Byogaichu Kenkyukaiho* (*Kyushu Pl. Prot. Res.*), **58**, 104-109 [In Japanese with English summary].
7. Li, X. Z. & Liu, Y. H. (2004) The biological characteristics and artificial rearing of *Cicadulina bipunctella* (Mots). *J. Southwest Agric. Univ.*, **26**, 143-145.
8. Li, X. et al. (2004) Effects of six host plants on the development and fecundity of *Cicadulina bipunctella*. *Chin. J. Appl. Ecol.*, **15**, 1431-1434 [In Chinese with English summary].
9. Lu, Z. X. et al. (2004) Effect of plant nitrogen on ecological fitness of the brown planthopper, *Nilaparvata lugens* Stål, in rice. *J. Asia- Pacific Entomol.*, **7**, 97-104.
10. Maramorosch, K. et al. (1961) Further studies on the maize and rice leaf galls induced by *Cicadulina bipunctella*. *Ent. Exp. et Appl.*, **4**, 86-89.
11. Matsukura, K. et al. (2009) Distribution, host plants, and seasonal occurrence of the maize orange leafhopper, *Cicadulina bipunctata* (Melichar) (Homoptera: Cicadellidae), in Japan. *Appl. Entomol. Zool.*, **44**, 207-214.
12. Matsukura, K. et al. (2009) Host manipulation by the orange leafhopper *Cicadulina bipunctata*: gall induction on distant leaves by dose-dependent stimulation. *Naturwissenschaften*, **96**, 1059-1066.
13. Matsukura, K. & Matsumura, M. (2009) Effect of infestation by the maize orange leafhopper *Cicadulina bipunctata* on six summer-seeding forage crop. *Kyushu Byogaichu Kenkyukaiho* (*Kyushu Pl. Prot. Res.*), **55**, 105-110 [In Japanese with English summary].
14. Matsukura, K. et al. (2010) Both nymphs and adults of the maize orange leafhopper induce galls on their host plant. *Commun. Integr. Biol.*, **3**, 388-389.
15. Matsukura, K. & Matsumura, M. (2010) Cultural control of leafhopper-induced maize wallaby-ear symptom in forage maize via early planting dates. *Crop Prot.*, **29**, 1401-1405.
16. Matsukura, K. et al. (2011) Efficient monitoring of orange leafhopper, *Cicadulina bipunctata* (Hemiptera: Cicadellidae) and small brown planthopper, *Laodelphax striatellus* (Hemiptera: Delphacidae), in forage maize fields using yellow sticky traps. *Appl. Entomol. Zool.*, **46**, 585-591.
17. Matsukura, K. et al. (2012) Estimation of climatic factors relating to occurrence of the maize orange leafhopper, *Cicadulina bipunctata*. *Popul. Ecol.*, **54**, 397-403.
18. Matsukura, K. et al. (2012) Host feeding by an herbivore improves the performance of offspring. *Evol. Biol.*, **39**, 341-347.
19. Matsukura, K. et al. (2013) Potential occurrence of wallaby ear symptom on forage crops in the five cropping in two years. *Nihon Oyo Dobutsu Konchu Gakkaishi* (*Jpn. J. Appl. Entomol. Zool.*), **57**, 11-18 [In Japanese with English summary].
20. Matsumura, M. et al. (2006) Recent outbreaks of the maize orange leafhopper *Cicadulina bipunctata* inducing gall-like structures on maize in Japan. *In* Gall arthropods and their associates: Ecology and evolution, eds. Ozaki, K.et al., Springer-Verlag, Tokyo, 149-158.
21. Matsumura, S. (1914) Die Jassinen und einige neue Acocephalinen Japans. *J. Coll. Agric. Tohoku Imp. Univ.*, **5**, 165-240 [In German].
22. Ofori, F. A. & Francki, R. I. B. (1983) Evidence that maize wallaby ear disease is caused by an insect toxin. *Ann. Appl. Biol.*, **103**, 185-189.
23. Ohata, S. (1993) The occurrence of the maize wallaby ear disease caused by the feeding of leafhopper, *Cicadulina bipunctata* Melichar (Homoptera: Cicadelidae). *Nihon Sochi Gakkaishi* (*Jpn. J. Grassl. Sci.*), **39**, 120-123 [In Japanese].
24. Tokuda, M. & Matsumura, M. (2005) Effect of temperature on the development and reproduction of the maize orange leafhopper *Cicadulina bipunctata* (Melichar) (Homoptera: Cicadellidae). *Appl. Entomol. Zool.*, **40**, 213-220.
25. Webb, M. D. (1987) Species recognition in *Cicadulina* leafhoppers (Homoptera: Cicadellidae), vectors of pathogens of Gramineae. *Bull. Entomol. Res.*, **77**, 683-712.
26. Weibull, J. & Melin, G. (1990) Free amino acid content of phloem sap from Brassica plants in relation to performance of *Lipaphis erysimi* (Hemiptera: Aphididae). *Ann. Appl. Biol.*, **116**, 417-423.