The Great Lakes Entomologist

Volume 38
Numbers 1 & 2 - Spring/Summer 2005 *Numbers 1 & 2 - Spring/Summer 2005*

Article 3

April 2005

Observations on the Overwintering Potential of the Striped Cucumber Beetle (Coleoptera: Chrysomelidae) in Southern Minnesota

R. L. Koch
University of Minnesota

M. A. Carrillo
University of Minnesota

E. C. Burkness
University of Minnesota

W. D. Hutchison
University of Minnesota

OBSERVATIONS ON THE OVERWINTERING POTENTIAL OF THE STRIPED CUCUMBER BEETLE (COLEOPTERA: CHRYSOMELIDAE) IN SOUTHERN MINNESOTA

R. L. Koch[1], M. A. Carrillo[1], E. C. Burkness[1], and W. D. Hutchison[1]

ABSTRACT

The striped cucumber beetle, *Acalymma vittatum* (Fabricius) (Coleoptera: Chrysomelidae), is an important pest of cucurbit crops. However, the overwintering capacity of this pest in temperate regions is poorly understood. In this study, the in-field survival of *A. vittatum* was examined during three consecutive winters. In addition, the supercooling points of *A. vittatum* were determined as an index of cold hardiness for adults. During each winter, the survival of adults decreased significantly through time, with no individuals surviving until spring. By comparing the supercooling points and in-field survival of adults to soil temperatures, it appears that winter temperatures in Minnesota are cold enough to induce freezing of the beetles. Moreover, a considerable amount of mortality occurred before minimum monthly soil temperatures dropped below the supercooling point of overwintering individuals, suggesting the occurrence of prefreeze mortality. An improved understanding of the response of *A. vittatum* to winter temperatures in temperate regions may aid in early season management of this pest.

INTRODUCTION

In Minnesota, USA, cucurbits (i.e., pumpkins, squash, cucumbers, and melons) are grown on 1,942 ha for a total value of $8,477,000 (Hutchison and O'Rourke 2003). The striped cucumber beetle, *Acalymma vittatum* (Fabricius) (Coleoptera: Chrysomelidae), is the most important insect pest of Minnesota cucurbits (Noetzel et al. 1985). Damage caused by this beetle results from adult and larval feeding injury and from transmission of pathogens (e.g., bacterial wilt, *Erwinia tracheiphila* [Smith]) (Capinera 2001). A diversity of research has been conducted to develop and improve integrated pest management (IPM) programs for *A. vittatum*, including sampling plans (Burkness and Hutchison 1997, Burkness and Hutchison 1998a), economic thresholds (Burkness and Hutchison 1998b, Brust and Foster 1999), biological control (Ellers-Kirk et al. 2000), and chemical control (Foster and Brust 1995, Brust et al. 1996). In temperate regions these IPM programs for *A. vittatum*, as with other pests, could benefit from an understanding of the capacity of the pest to survive winter conditions.

The magnitude of early season pest infestations can depend upon the overwintering survival of the pest, which relates primarily to its cold hardiness (Leather et al. 1993). Insects that survive freezing temperatures are generally classified under two strategies of survival, freeze tolerance or freeze intolerance (Salt 1961). Freeze-tolerant insects survive extracellular ice formation, while freeze-intolerant insects avoid ice formation by supercooling or by seeking physical protection from their microhabitat (e.g., Lee Jr. and Denlinger 1991, Leather et al. 1993). The supercooling point, defined as the temperature at which body fluids spontaneously freeze (Zachariassen 1985), represents the absolute lower

[1]Department of Entomology, 219 Hodson Hall, 1980 Folwell Avenue, University of Minnesota, St. Paul, Minnesota, 55108, USA.

lethal temperature for freeze-intolerant individuals (Lee Jr. and Denlinger 1991). However, death may occur at warmer temperatures due to chill injury (Baust and Rojas 1985, Lee Jr. and Denlinger 1985, Knight et al. 1986, Bale 1996). For example, aphids have relatively low supercooling points, but generally attain high mortality at temperatures well above the supercooling point (Knight et al. 1986). Regardless, the supercooling point is often measured in cold hardiness studies to aid in classifying the survival strategy at low temperatures (e.g., Lee Jr. and Denlinger 1991, Renault et al. 2002, Koch et al. 2004, Carrillo et al. 2005a).

To our knowledge, the overwintering potential of *A. vittatum* has not been examined. However, in other U.S. locations it has been observed that *A. vittatum* adults overwinter under debris (Cranshaw 2004) in woodlands, hedgerows, and weedy fence rows (Houser and Balduf 1925, Gould 1944). The objective of this study was to examine the overwintering survival of adult *A. vittatum* in southern Minnesota. In addition, the supercooling point was measured as an index of cold hardiness.

MATERIALS AND METHODS

Overwintering survival. In-field survivorship experiments were conducted in the winters of 2002-2003, 2003-2004, and 2004-2005. On 17 September 2002, three cages were placed in a wooded area at the University of Minnesota Outreach, Research, and Education (UMORE) Park, Rosemount, Minnesota. Each cage consisted of a 45 × 45 × 70 cm wooden frame covered with fine nylon mesh on the top and sides. The bottom of each cage was open and the sides were buried 5 cm into the soil. The mesh on the top of the cage was attached with Velcro, so that the cage could be opened. A ca. 5 cm layer of deciduous leaf litter and grass was placed on the soil surface in each cage. Fifty *A. vittatum* adults (ca. 7 mm long each, Capinera 2001) collected on the same date from a squash field at UMORE Park were released into each cage. In addition, a piece of 'We-Be-Little' pumpkin (6 × 6 × 4 cm) was placed into each cage to provide the beetles with food. On 20 September 2002, 30 additional *A. vittatum* were added to each cage, resulting in a total of 80 beetles per cage. On 9 April 2003, the cages were opened, and the leaf litter and top 2.5 cm of soil were carefully examined for surviving beetles. Survival was defined as the presence of movement after beetles were prodded with a pair of soft forceps.

For the winters of 2003-2004 and 2004-2005, cages consisted of plastic dishes (8.0 cm high × 18.5 cm diameter; Pioneer Plastics, Inc., Dixon, KY) with a 10.2-cm diameter hole cut into the bottom of the dish and the lid to provide ventilation and drainage. The holes were covered with fine, 30-mesh wire cloth (The Cleveland Wire Cloth and Mfg. Co., Cleveland, OH), which was attached to the dishes using silicone sealant (GE Sealants and Adhesives, Huntersville, NC). On 6 October 2003, 11 cages were placed into a wooded area at UMORE Park. A 3.8-cm-thick plug of sod (soil with roots and above ground growth) containing deciduous leaf litter was placed into each cage, with the sod cut to cover the entire bottom of the cage. In addition, a piece of a 'We-Be-Little' pumpkin (3 × 3 × 2 cm) was also placed on top of the sod in each cage as a food source for the beetles. The cages were then buried in the soil so that the top of the soil in each cage was level with that outside the cage. Thirty *A. vittatum* adults were collected on the same date from a squash field at UMORE Park, and were placed into each cage. The lids were placed on the cages, and the cages were covered with a 3-cm layer of deciduous leaf litter. Throughout the winter, cages were retrieved from the field and returned to the laboratory, with two cages being retrieved on each of 20 November, 15 December 2003, 13 January, 16 February, and 15 March 2004, and one cage on 15 April 2004. In the laboratory, the cages were maintained at 25 ºC for 24 h, and then thoroughly inspected for surviving *A. vittatum*, with survival defined as previously explained.

On 19 October 2004, eight cages were constructed and deployed in the field as described for the 2003-2004 winter. Each cage contained 30 adult *A. vittatum* collected from a squash field at UMORE Park. Throughout the winter, cages were retrieved from the field and brought back to the laboratory, with two cages retrieved on each of 18 November, 15 December 2004, 19 January, and 8 February 2005. Survival was assessed in the laboratory, as previously explained, after the cages were held at 25 ºC for 24 h.

The proportionate survival after initiation of the experiment for each cage was arcsine-square-root transformed (Southwood and Henderson 2000), and the data were analyzed separately for each winter using analysis of variance (ANOVA) (PROC GLM, SAS Institute 1995). Month within each winter was the only predictor included in the ANOVA models. Means were separated using the REGWQ test at $\alpha = 0.05$.

Supercooling point. Adult *A. vittatum* were collected at UMORE Park and returned to the laboratory for supercooling point measurements. In 2003 beetles were collected using an aspirator on 21 November from a cucurbit field. An additional supercooling point measurement was taken on surviving beetles retrieved from the survivorship study on 15 December 2003. In 2004, supercooling point measurements were made on surviving beetles retrieved from the survivorship study on 19 November and 15 December.

Following the methods of Carrillo et al. (2004), supercooling points were determined, using surface-contact thermometry, as an index of cold hardiness. Adult *A. vittatum* were attached to 24-gauge copper-constantan thermocouples using a thin layer of high-vacuum grease (Dow Corning®, Dow Corning Corporation, Midland, MI), and placed at the center of 19 × 19 × 19 cm polystyrene containers with a starting temperature of ca. 0 ºC. Prior to thermocouple attachment, individuals were immobilized at ca. 0 ºC for 2 min. Polystyrene containers were closed with rubber stoppers, and then transferred to -80 ºC in an ultra-low-temperature freezer (Revco Scientific Inc., Asheville, NC) to cool the beetles at ca. 1 ºC min^{-1} (Carrillo et al. 2004). For each cooling period, a total of five containers were used with two beetles per container. During cooling, the temperature of each individual was recorded with a multi-channel data logger (Personal Daq/56 data acquisition system, IOtech, Inc., Cleveland, OH) that transferred data at 1-s intervals through a USB cable into a desktop computer. Temperature data were downloaded to a spreadsheet (Microsoft Excel) and graphed as a scatter plot. On the scatter plot, the supercooling point was identified as the lowest temperature attained prior to the sudden increase in temperature, indicative of the heat released during the phase change from liquid to solid (Lee Jr. 1991).

In addition, supercooling points and adult survival from the study described above were compared to soil temperatures measured 2.5 cm below the surface 20 cm away from the cages used in the survivorship study at UMORE Park. Temperature data were recorded using a HOBO® data logger (Onset Computer Corporation, Cape Cod, MA).

RESULTS AND DISCUSSION

In Minnesota, it has remained uncertain whether *A. vittatum* successfully overwinters or if it immigrates into the state each year (Burkness 1996). In the preliminary in-field survivorship study conducted in the winter of 2002-2003, no beetles survived through the winter (Fig. 1A). In more detailed in-field survivorship studies conducted in the winters of 2003-2004 and 2004-2005, *A. vittatum* survival decreased significantly as winter progressed (2003-2004: $F = 19.51$; df = 5, 5; $P = 0.0027$; 2004-2005: $F = 21.02$; df = 3, 4; $P = 0.0065$), with 0% survival of adults by spring (Fig. 1BC). These results contrast those obtained for adult bean leaf beetle, *Cerotoma trifurcata* (Förster) (Coleoptera: Chrysomelidae)

(Carrillo et al. 2005b), which has a similar overwintering behavior (Lam and Pedigo 2004). Under the studied conditions, it seems unlikely that *A. vittatum* consistently overwinters in Minnesota. However, further research is needed to determine the causes of *A. vittatum* mortality at low temperatures under varying field conditions. In addition to temperature, the effect of other factors on beetle mortality should be evaluated. For example, Lam and Pedigo (2000) examined the potential impact of entomopathogenic fungi and ectoparasitic mites on overwintering *C. trifurcata*. Furthermore, the impact of abiotic factors (e.g., moisture) on overwintering mortality should be studied.

Soil temperatures at the experimental sites were low enough to induce freezing in all individuals. The mean supercooling points recorded at the beginning of the winters were -5.5 ºC in December 2003 and -6.5 ºC in December 2004 (Table 1). If we assume the average of these two values (i.e., -6.0 ºC) to be the overwintering supercooling point for this species, we find that winter minimum monthly soil temperatures dropped below this value in all years (Fig. 1), suggesting that *A. vittatum* adults could have frozen. However, it is interesting to note that temperatures dropped below the overwintering supercooling point after a considerable amount of mortality had already occurred, indicating mortality due to factors other than freezing (Lee Jr. and Denlinger 1985, Lee Jr. and Denlinger 1991). A direct comparison between the supercooling point and mortality at different subzero temperatures under controlled conditions should be conducted to determine the strategy of survival (i.e., freeze tolerance or freeze intolerance) of *A. vittatum* during winter months (e.g., Bale 1991, Carrillo et al. 2005a). Such work would also elucidate the ecological relevance of the supercooling point as an index of cold hardiness for this pest.

To our knowledge, this study is the first report on the overwintering potential of *A. vittatum* in temperate regions. Our results suggest that *A. vittatum* is unlikely to overwinter in Minnesota. However, it is possible that some beetles may survive under microhabitat conditions not captured by our study. Therefore, initial infestations of Minnesota cucurbits may result primarily from immigration of *A. vittatum* from southern locations. The present results improve our understanding of the response of this pest to winter temperatures in temperate regions. Furthermore, results from this study may aid in forecasting early season populations (e.g., Carrillo et al. 2005b) as part of an overall IPM program for this pest.

Table 1. Late season supercooling points of adult *Acalymma vittatum* in southern Minnesota, USA.

		Supercooling point (°C)		
Month	**n**	**Mean ± SE**	**Median**	**Range**
		2003		
November	17	-6.0 ± 0.20	-6.2	-7.1, -3.9
December	13	-5.5 ± 0.41	-5.6	-7.5, -3.0
		2004		
November	21	-5.3 ± 0.23	-5.5	-7.3, -3.1
December	21	-6.5 ± 0.22	-6.4	-8.4, -4.1

ACKNOWLEDGEMENTS

We thank S. Wold-Burkness, T. Galvan, and C. Kulhanek (University of Minnesota) for field and laboratory assistance. This research was funded by University of Minnesota Doctoral Dissertation Fellowships awarded to R.L.K. and M.A.C., and the University of Minnesota Experiment Station.

LITERATURE CITED

Bale, J. S. 1991. Implications of cold hardiness for pest management, pp. 461-498. *In* R. E. Lee Jr., and D. L. Denlinger (eds.), Insects at Low Temperature. Chapman and Hall, New York.

Bale, J. S. 1996. Insect cold hardiness: A matter of life and death. Eur. J. Entomol. 93: 369-382.

Baust, J. G., and R. R. Rojas. 1985. Insect cold hardiness: Facts and fancy. A review. J. Insect Physiol. 31: 755-759.

Brust, G. E., R. E. Foster, and W. G. Buhler. 1996. Comparison of insecticide use programs for managing the striped cucumber beetle (Coleoptera, Chrysomelidae) in muskmelon. J. Econ. Entomol. 89: 981-986.

Brust, G. E., and R. E. Foster. 1999. New economic threshold for striped cucumber beetle (Coleoptera: Chrysomelidae) in cantaloupe in the Midwest. J. Econ. Entomol. 92: 936-940.

Burkness, E. C. 1996. Action thresholds and sequential sampling plans for the striped cucumber beetle (Coleoptera: Chrysomelidae) in cucurbits. M. S. Thesis, University of Minnesota. St. Paul, MN.

Burkness, E. C., and W. D. Hutchison. 1997. Development and validation of a binomial sequential sampling plan for striped cucumber beetle (Coleoptera: Chrysomelidae) in cucurbits. J. Econ. Entomol. 90: 1590-1594.

Burkness, E. C., and W. D. Hutchison. 1998a. Development and validation of a fixed-precision sampling plan for estimating striped cucumber beetle (Coleoptera: Chrysomelidae) density in cucurbits. Environ. Entomol. 27: 178-183.

Burkness, E. C., and W. D. Hutchison. 1998b. Action thresholds for striped cucumber beetle (Coleoptera: Chrysomelidae) on Carolina cucumber. Crop Prot. 17: 331-336.

Capinera, J. L. 2001. Handbook of Vegetable Pests. Academic Press, San Diego.

Carrillo, M. A., N. Kaliyan, C. A. Cannon, R. V. Morey, and W. F. Wilcke. 2004. A simple method to adjust cooling rates for supercooling point determination. CryoLetters 25: 155-160.

Carrillo, M. A., C. A. Cannon, W. F. Wilcke, R. V. Morey, N. Kaliyan, and W. D. Hutchison. 2005a. Relationship between supercooling point and mortality at low temperatures in Indianmeal moth (Lepidoptera: Pyralidae). J. Econ. Entomol. 98: 618-625.

Carrillo, M. A., R. L. Koch, E. C. Burkness, K. Bennett, D. W. Ragsdale, and W. D. Hutchison. 2005b. Supercooling point of bean leaf beetle (Coleoptera: Chrysomelidae) in Minnesota and a revised predictive model for survival at low temperatures. Environ. Entomol. 34: 1395-1401.

Cranshaw, W. 2004. Garden Insects of North America. Princeton University Press, Princeton.

Ellers-Kirk, C. D., S. J. Fleischer, R. H. Snyder, and J. P. Lynch. 2000. Potential of entomopathogenic nematodes for biological control of *Acalymma vittatum* (Coleoptera: Chrysomelidae) in cucumbers grown in conventional and organic soil management systems. J. Econ. Entomol. 93: 605-612.

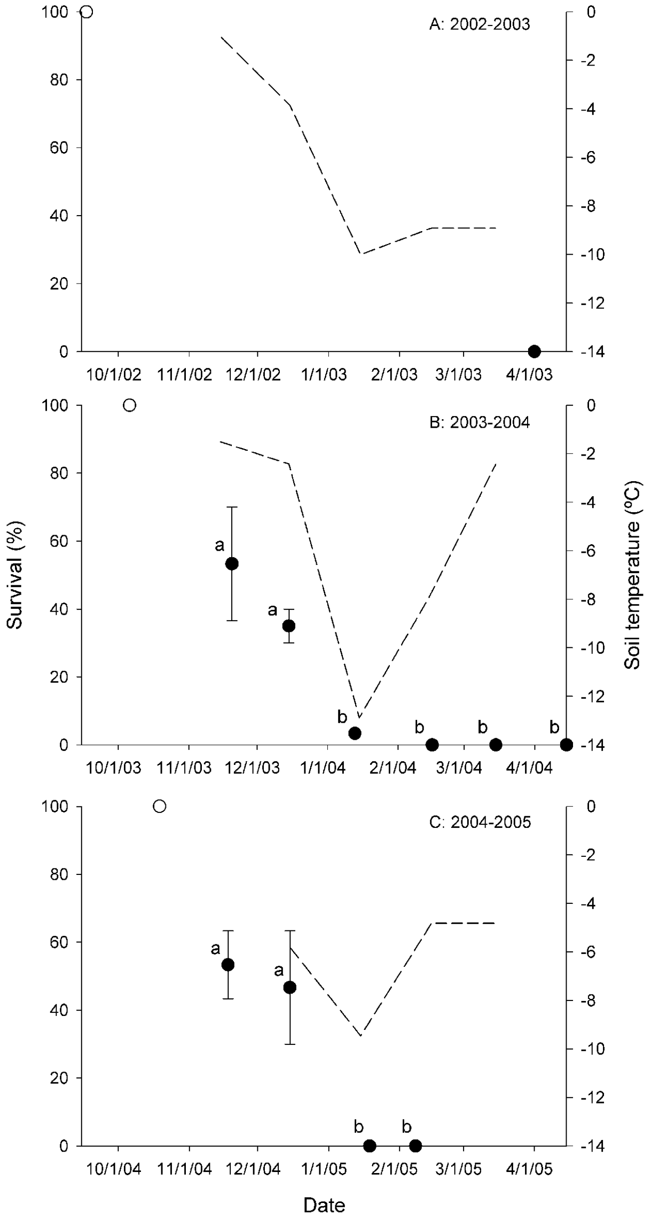

Figure 1. Mean (± SE) overwintering survival of *Acalymma vittatum* adults (circles) and minimum monthly soil temperature recorded 2.5 cm below the soil surface (dashed line) at Rosemount, Minnesota, USA. All trials started with 100% survival (open symbols), and the survival at subsequent sample dates is represented by filled symbols. Means within winters with different letters differ significantly ($P < 0.05$; analysis of variance and REGWQ test). The overwintering mean supercooling point of this insect ranged between -6.5 and -5.3 ºC among the years studied.

Foster, R. E., and G. E. Brust. 1995. Effects of insecticides applied to control cucumber beetles (Coleoptera, Chrysomelidae) on watermelon yields. Crop Prot. 14: 619-624.

Gould, G. E. 1944. The biology and control of the striped cucumber beetle. Purdue University Agricultural Experiment Station Bulletin 490.

Houser, J. S., and W. V. Balduf. 1925. The striped cucumber beetle. Ohio Agricultural Experiment Station. Bulletin 388.

Hutchison, W. D., and P. K. O'Rourke. 2003. Annual Production and Value for Major Vegetable and Fruit Crops in Minnesota. MN Project, St. Paul, MN (http://pestmanagementcenter-mn.coafes.umn.edu/mnpiap/MN%20Production%20and%20Ag%20Stats.pdf).

Knight, J. D., J. S. Bale, F. Franks, S. F. Mathias, and J. G. Baust. 1986. Insect cold hardiness – Supercooling points and prefreeze mortality. Cryo-Letters 7: 194-203.

Koch, R. L., M. A. Carrillo, R. C. Venette, C. A. Cannon, and W. D. Hutchison. 2004. Cold hardiness of the multicolored Asian lady beetle (Coleoptera: Coccinellidae). Environ. Entomol. 33: 815-822.

Lam, W. K. F., and L. P. Pedigo. 2000. A predictive model for the survival of overwintering bean leaf beetles (Coleoptera: Chrysomelidae). Environ. Entomol. 29: 800-806.

Lam, W. K. F., and L. P. Pedigo. 2004. Ecology and management of the bean leaf beetle *Cerotoma trifurcata*, pp. 579-589. *In* P. Jolivet, J. A. Santiago-Blay, and M. Schmitt (eds.), New Developments in the Biology of Chrysomelidae. Academic Publishing, The Hague.

Leather, S. R., K. F. A. Walters, and J. S. Bale. 1993. The Ecology of Insect Overwintering. Cambridge University Press, Cambridge.

Lee, R. E., Jr., and D. L. Denlinger. 1985. Cold tolerance in diapausing and non-diapausing stages of the flesh fly, *Sarcophaga crassipalpis*. Physiol. Entomol. 10: 309-315.

Lee, R. E. Jr., and D. L., Denlinger. 1991. Insects at Low Temperature. Chapman and Hall, New York.

Noetzel, D. M., L. K. Cutkomp, and P. K. Harein. 1985. Estimated Annual Losses Due to Insects in Minnesota 1981-1983. University of Minnesota. Agricultural Extension Service, AG-BU-2541.

Renault, D., C. Salin, G. Vannier, and P. Vernon. 2002. Survival at low temperatures in insects: What is the ecological significance of the supercooling point? CryoLetters 23: 217-228.

Salt, R. W. 1961. Principles of insect cold-hardiness. Ann. Rev. Entomol. 6: 55-74.

SAS Institute. 1995. User's Manual, version 6, 4th edition. SAS Institute, Cary, NC.

Southwood, T. R. E., P. A. Henderson. 2000. Ecological Methods. Blackwell Ltd., Oxford.

Zachariassen, K. E. 1985. Physiology of cold tolerance in insects. Physiol. Revs. 65: 799-832.