BioOne COMPLETE

FEEDING EFFECTS OF ISCHNODEMUS VARIEGATUS (HEMIPTERA: BLISSIDAE) ON PHOTOSYNTHESIS AND GROWTH OF HYMENACHNE AMPLEXICAULIS (POACEAE)

Authors: Overholt, William A., Ewe, Sharon M. L., Diaz, Rodrigo, Morgan, Eric C., and Moeri, Onour E.

FEEDING EFFECTS OF *ISCHNODEMUS VARIEGATUS* (HEMIPTERA: BLISSIDAE) ON PHOTOSYNTHESIS AND GROWTH OF *HYMENACHNE AMPLEXICAULIS* (POACEAE)

William A. Overholt[1], Sharon M. L. Ewe[2], Rodrigo Diaz[1], Eric C. Morgan[1] and Onour E. Moeri[1]
[1]University of Florida, IFAS, 2199 South Rock Road, Fort Pierce, FL 34945

[2]Florida International University, Southeast Research Center, Miami, FL 33199

Abstract

The influence of *Ischnodemus variegatus* feeding on photosynthesis and growth of the invasive semi-aquatic grass, *Hymenachne amplexicaulis*, was investigated in field and greenhouse environments. In the field, carbon dioxide assimilation of infested plants was approximately 35% less than that of non-infested plants, and the rate of assimilation was related to *I. variegatus* density. The relative growth rate of infested plants in the greenhouse was 77% of that of non-infested plants, and biomass of infested plants was significantly less than for non-infested plants 79 days after infestation. The value of *I. variegatus* as a fortuitous biological control agent of *H. amplexicaulis* is discussed.

Key Words: invasive plants, biological control, photosynthesis, wetlands.

Resumen

La influencia de la alimentación de *Ischnodemus variegatus* sobre la fotosíntesis y el desarrollo del pasto invasivo semi-acuatico *Hymenachne amplexicaulis*, fue investigado en ambientes de campo e invernadero. En el campo, la asimilación del dióxido de carbono por plantas infestadas fue aproximadamente 35% menos que en las plantas no infestadas y la tasa de asimilación fue relacionada con la densidad de *I. variegatus*. La tasa de crecimiento relativo de plantas infestadas en el invernadero fue 77% menos que las plantas no infestadas y la biomasa de plantas infestadas fue significativamente menos en plantas no infestadas a los 79 días después de la infestación. Se discute sobre el valor de *I. variegatus* como un agente de control biológico fortuito.

Hymenachne amplexicaulis Rudge (Nees) (Poaceae), commonly referred to as West Indian Marsh Grass, is an exotic, perennial, semi-aquatic grass which was first seen in Florida in 1957 (University of Florida Herbarium 2003). The native range of the grass is tropical Central and South America (Bogman 1977). *Hymenachne amplexicaulis* reproduces and grows from stolons or seeds in areas with fluctuating water levels. It can survive long periods of flooding, but will only persist along the edges of permanent deep water (Tejos 1980). Csurhes et al. (1999) stated that the plant preferred low lying fresh water wetlands and flood plains, and grew most prolifically in wetlands which receive high nutrient and sediment influx.

In the 1970s and 80s, *H. amplexicaulis* began invading wetlands in southern and central Florida (Langeland & Craddock-Burks 1998). The Florida Exotic Pest Plant Council has listed *H. amplexicaulis* as a Category I invasive plant (FLEPPC 2003), and it is included on the Florida Department of Environmental Protection's list of noxious plants (DEP 2003). Although no quantitative studies have yet been conducted to examine the effect of *H. amplexicaulis* on wetland biodiversity, it has clearly displaced native plant species in some areas, particularly in marshes in Myakka River State Park where large monocultures of the grass occur (Langeland & Craddock-Burks 1998; R. Diaz unpublished data).

In 2000, a biologist at Myakka River State Park noticed an insect causing considerable damage to *H. amplexicaulis* in the park (P. Benshoff, Park Naturalist, Myakka River State Park, pers. comm.). Specimens of the insect sent to the Florida Department of Agriculture and Consumer Services were identified as *Ischnodemus variegatus* Signoret (Hemiptera: Blissidae), a new record for Florida (Halbert 2000). Previously, *I. variegatus* had been reported from several countries in Central and South America (Baranowski 1979; Slater 1987). *Hymenachne amplexicaulis* is the only host mentioned for *I. variegatus* in South America, although Baranowski (1979) cites a 'sitting' record on *Thalia geniculata* L. (Marantaceae) from Suriname. The objective of the present study was to quantify the effect of *I. variegatus* feeding on photosynthesis and growth of *H. amplexicaulis*.

MATERIALS AND METHODS

Field Measurements

A portable infra-red gas exchange system (CIRAS-1, PP Systems, Massachusetts, USA) was used to measure leaf photosynthesis (i.e., net carbon dioxide (CO_2) assimilation). Gas exchange was measured on plants growing along the banks of Fisheating Creek (Glades Co.) (26.95°N, 81.14°W) on the western side of Lake Okeechobee on three days (28/8, 4/9, 18/9) during August and September 2003. On each sampling date, photosynthesis was measured on 27-66 *H. amplexicaulis* plants. In order to increase the chance of having both infested and non-infested plants in the sample, plants were selected according to leaf color. Previous observations (W. Overholt, unpublished data) on laboratory infested plants indicated that feeding by *I. variegatus* induced a change in color of *H. amplexicaulis* leaves from green to dark red. On each sampling date, an approximately equal number of plants were sampled from patches of *H. amplexicaulis* showing no damage symptoms and patches with red leaves. After measuring gas exchange, culms were excised at water level and dissected to count nymphs and adults of *I. variegatus*.

Plants

Stolons and seeds of *H. amplexicaulis* were collected in Myakka River State Park from October to December, 2002, and used to propagate plants in a greenhouse in Fort Pierce. Plants were grown in 1-L plastic pots in commercial potting soil from rooted stolon cuttings or seeds. Pots were placed in solid-bottomed trays, to which water was added and maintained at a depth of 4-6 cm.

Insects

Adults and nymphs of *I. variegatus* were collected in Myakka River State Park and taken to the laboratory. *Hymenachne amplexicaulis* plants (~30 cm in height) were inoculated with 10 *I. variegatus* adults/nymphs. Plants in pots were placed in trays with water, and maintained inside a PVC framed cage covered with fine organdy cloth.

Greenhouse Bioassay

The bioassay was conducted under ambient conditions (22.2-37.2°C, mean temperature of 27.1°C, natural lighting) in a greenhouse at the University of Florida's Indian River Research and Education Center in Fort Pierce. Plants used for the bioassay were grown from seed planted on the same day, and grown under the same conditions for 80 days. All plants were grown in 1-L plastic pots containing commercial potting soil, and at the time of infestation were approximately the same size. Six plants were randomly selected from this group, and infested with 20 *I. variegatus* second instars, and six plants were selected as non-infested controls. In addition, six plants were removed from their pots, dried, and weighed to estimate initial biomass. Infested and non-infested plants were caged in open-bottomed acrylic cylinders (46 cm × 14.5 cm, height × diameter) in which the top was covered with 300 μ mesh nylon cloth. Holes (8.5 cm diameter) were cut at 6 locations in the sides of the tubes and replaced with the same nylon cloth to allow ventilation. Plants and their cages were placed in trays and water was added to a depth of 4-6 cm. Water was replenished daily to maintain this depth throughout the course of the experiment.

Net CO_2 assimilation was measured once or twice a week on the second fully expanded leaf from the top of the plant with the CIRAS-1 instrument. The first measurements were taken just prior to infestation, and the last measurements were made at 79 days after infestation. After 79 days, infested plants were dissected and all *I. variegatus* individuals removed, classified by age (nymphal instars 1-5 or adult), and counted. Eggs were difficult to locate due to their small size and cryptic coloration, and were not counted.

At harvest, the numbers of leaves and culms were counted. Basal diameter of each culm and the length and width of the largest leaf of each plant were measured with a micrometer. All growth medium was then washed off the plants. Leaves were separated from the roots and all leaves digitally photographed. Leaf area was determined from the digital images with ImageJ software (ImageJ shareware, NIH, Bethesda, MD). The proportion of damaged area also was assessed by this method. Plants were then dried in an oven at 65°C for two weeks and weighed to obtain measurements of biomass.

Data Analysis

CO_2 assimilation in the field was analyzed with two-way analysis-of-covariance (ANCOVA) with date and infestation status (infested vs. non-infested, where infested plants had one or more nymphs and/or adults of *I. variegatus*) as main effects and light level as a covariate (PROC GLM, SAS Institute 2001). The covariate was included in the model because light intensity varied greatly among observations, ranging from 502 to 1964 μmol photons $m^{-2}s^{-1}$. The number of *I. variegatus* was also regressed on CO_2 assimilation (PROC REG, SAS Institute 2001). Greenhouse gas exchange was analyzed with repeated measures analysis-of-variance (ANOVA) (PROC GLM, SAS Institute, 2001). We used *t*-tests to compare leaf area, morphometric parameters and growth rates between infested and control plants.

RESULTS

Field Measurements

Mean CO_2 assimilation was different between sampling dates ($F = 53.1$, $df = 2, 132$, $P < 0.0001$) and between infested plants (8.5 ± 0.5 µmol CO_2 m^{-2}s^{-1}) and non-infested plants (12.9 ± 0.6 µmol CO_2 m^{-2}s^{-1}) ($F = 42.3$, $df = 4, 132$, $P < 0.0001$). The covariate light was also significant ($F = 5.12$, $df = 1, 132$, $P < 0.025$). A regression of CO_2 assimilation on number of insects showed a decline in photosynthesis as insect numbers increased ($F = 21.5$, $df = 1, 135$, $P < 0.0001$, $R^2 = 0.13$) (Fig. 1).

Greenhouse Bioassay

The number of *I. variegatus* found on plants at harvest ranged from 50 to 149, with a mean of 97 ± 13 (SE). Approximately 86% of the individuals removed from the plants were either first (70.1%) or second (15.9%) instars. As the plants were infested with 20 second instars, this represented a population increase of about 2.5 to 7.5 times during the 79-day experiment. A generation of *I. variegatus* is completed in approximately 60 days (R. Diaz, unpublished), and therefore, the insects found at harvest were most likely the F$_1$ progeny of those released.

Growth patterns of the infested plants were different from those of the control plants (Fig. 2, Table 1). The amount of damage, as quantified by the amount of red leaf area, in the infested plants was higher ($F = 4.722$, $df = 1, 10$, $P < 0.01$) than non-infested plants (Table 1).

The relative growth rate of infested plants was 77% that of the control plants, which resulted in
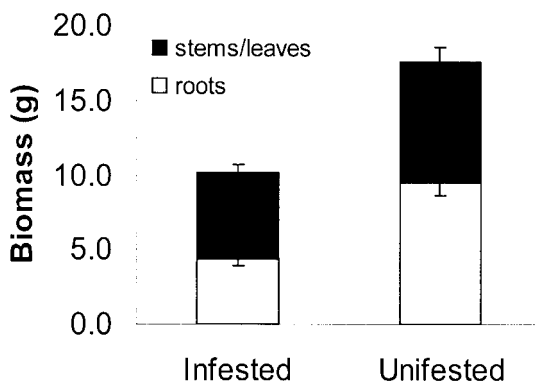

Figure 2. Average biomass (± SEM) allocated to roots and shoots for treatment and control plants.

much larger control plants (Table 1). Both root and shoot biomass were greater in the controls relative to the infested plants (roots: $F = 3.437$, $df = 1,10$, $P < 0.01$; shoots: $F = 1.968$, $df = 1,10$, $P < 0.05$) (Fig. 2). On average, control plants were more than 1.5 times larger than the infested plants. Neither infested nor non-infested plants flowered during the experiment, and no plants died.

Morphometric differences were observed between the two treatments. Control plants had larger total leaf area, longer leaves and thicker culms than the infested individuals (Table 1). The number of culms, however, was not different between treatments.

Carbon dioxide assimilation rates were higher in the control plants during the first seven sampling periods (days 4-29), but were not different after day 29 (Fig. 3). Repeated measures ANOVA indicated a difference between treatments ($F = 6.92$, $df = 1,10$, $P < 0.05$, power = 0.660).

DISCUSSION

Feeding by *Ischnodemus variegatus* adults and nymphs clearly affected the overall growth of *H. amplexicaulis*. Both above and below ground parts of infested plants were smaller than those of uninfested plants. Infested plants also had a higher proportion of damaged leaves relative to the controls. In addition to the effect of infestation on biomass, the majority of leaves of infested plants were red or brown at harvest. The reddish discoloration of the leaves, suggestive of a breakdown and/or disruption in the function of plant photosynthetic pigment complexes, is similar to damage symptoms on corn (*Zea mays* L.) and buffalograss (*Buchloë dactyloides* (Nuttall)) caused by feeding of *Blissus* spp. (Hemiptera: Blissidae) (Negron & Riley 1990; Baxendale et al. 1999).

In the greenhouse bioassay, CO_2 assimilation was lower in infested plants for the first 29 days after infestation. It was somewhat surprising
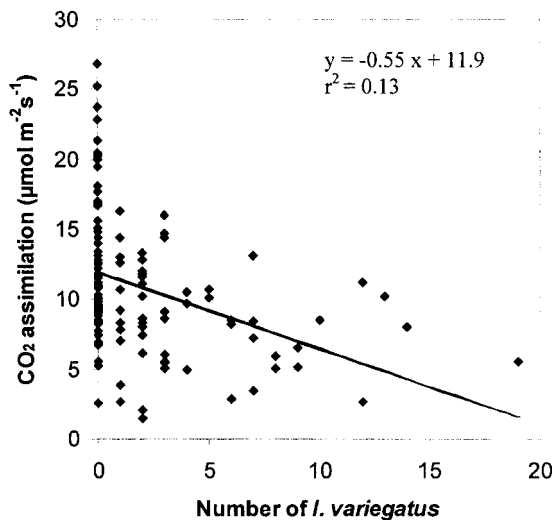

Figure 1. Carbon dioxide assimilation of plants in the field with variable densities of *I. variegatus*.

$$y = -0.55x + 11.9$$
$$r^2 = 0.13$$

TABLE 1. AVERAGE (± SEM) VALUES OF MEASURED PARAMETERS IN PLANTS WITH *I. VARIEGATUS* AND CONTROL PLANTS. SIGNIFICANT *P*-VALUES FROM *T*-TEST COMPARISONS ARE SHOWN IN BOLD.

	Infested	Control	*P*-value
Leaf area (cm^2):			
Damaged	69 ± 9	17 ± 1	0.000
Undamaged	201 ± 30	441 ± 42	0.001
Total	270 ± 29	458 ± 43	0.005
Largest leaf (cm):			
Length	18.8 ± 0.7	22.3 ± 0.5	0.003
Width	1.2 ± 0.1	1.16 ± 0.0	0.429
Culm:			
Number	6.8 ± 0.4	8.5 ± 1.1	0.171
Thickness (mm)	3.4 ± 0.1	4.0 ± 0.1	0.002
Relative growth rate (g/g/week)[1]	0.167 ± 0.01	0.215 ± 0.01	0.008

[1]Grams biomass gained/grams initial plant biomass/week.

that CO_2 assimilation was not different between infested and non-infested plants from 32 days after infestation until the experiment was terminated on day 79. We suspect that the size of the acrylic tubes covering the plants may have negatively influenced growth. Both infested and non-infested plants grew to the top of the tubes within the first month, after which their growth may have been impaired by the size of the containers. The small chamber size may also explain the color changes in leaves of non-infested plants (Table 1).

Insect herbivores access plant resources either through consumption of foliage or other solid materials (chewing insects) or by ingesting plant sap (piercing/sucking insects). Piercing/sucking insects, such as blissids, feed in phloem, xylem, epidermal, or mesophyll paraenchyma tissues (Walling 2000). Xylem feeding is reported to inhibit photosynthesis more than other types of herbivory (Meyer & Whitlow 1992). Johnson & Knapp (1996) concluded that photosynthetic inhibition of *Spartina falicus* (Link) (Poaceae) caused by *Ischnodemus falicus* (Say) was consistent with xylem feeding. Our field and greenhouse results clearly demonstrated that *I. variegatus* decreased photosynthetic capacity of *H. amplexicaulis*. Although the tissues accessed during feeding were not identified, we are doubtful that this insect is a xylem feeder. Press & Whittaker (1993) stated that there is little evidence to support xylem feeding of insects other than cercopids, cicadids, and some cicadellids. Moreover, we saw no evidence of copious amounts of excreted liquids typically associated with xylem feeding (Press & Whittaker 1993).

Determining the value of *I. variegatus* as a biological control agent of *H. amplexicaulis* requires additional research. The insect undoubtedly affected photosynthesis and plant growth, but even at relatively high initial densities of 20 *I. variegatus* per plant, which increased to an average of 97 insects per plant during the course of the experiment, plants were not killed. Densities of *I. variegatus* monitored in the field in 2002/2003 rarely surpassed 20 insects per plant, and were most often much lower (Overholt, unpublished data). However, in the field, plants are stressed by a variety of factors, including climate, water, soil nutrient levels, pathogens, herbivores, and competition with other plants. *Hymenachne amplexicaulis* has low drought tolerance (Medina & Motta 1990) and thus a combined effect of low water availability and insect damage may have additive negative effects on *H. amplexicaulis* growth. Additionally, *I. variegatus* may influence flowering and seed production, which were not measured in our experiments. A negative impact on seed production could conceivably slow the spread of *H. amplexicaulis* in Florida's wetlands.
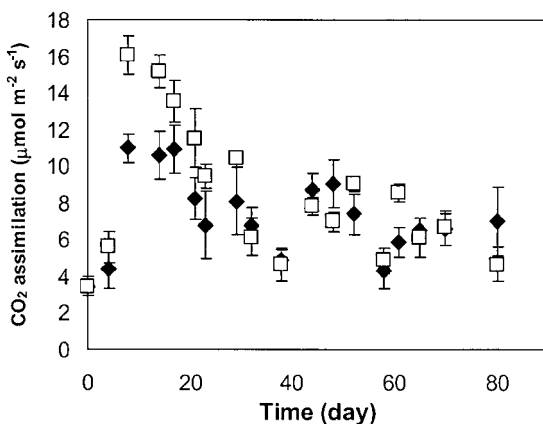

Figure 3. Average rates of carbon dioxide net assimilation (± SEM) in infested (◆) and control (□) plants for each sample day.

Finally, the value of *I. variegatus* to natural resource management and agriculture in Florida will depend on its host range. In South America, *I. variegatus* has been recorded only from *H. amplexicaulis* (Baranowski 1979), and there are no native members of this genus in Florida or the USA (reference). However, no detailed studies of the insect's biology have been conducted. We are currently investigating the host range of *I. variegatus* by measuring survivorship on a large number of native and economically important grasses. If *I. variegatus* is restricted to *Hymenachne*, there is little threat of its shifting to more distantly related grasses (Pemberton 2000).

REFERENCES CITED

BARANOWSKI, R. M. 1979. Notes on the biology of *Ischnodemus oblongus* and *I. fulvipes* with descriptions of the immature stages. Ann. Entomol. Soc. Amer. 72: 655-658.

BAXENDALE, F. P., T. M. HENG-MOSS, AND T. R. RIORDAN. 1999. *Blissus occiduus* (Hemiptera: Lygaeidae): a chinch bug pest new to buffalograss turf. J. Econ. Entomol. 92: 1172-1176.

BOGMAN, A. V. 1977. Tropical Pasture and Fodder Plants. Longman, New York. 475 pp.

CSURHES, S. M., A. P. MACKEY, AND L. FITZSIMMONS. 1999. *Hymenache* (*Hymenachne amplexicaulis*) in Queensland. Queensland Government, Department of Natural Resources and Mines. 39 pp. http://www.nrm.qld.gov.au/pests/psas/pdfs/Hymenachne.pdf

DEP. 2003. Florida Department of Environmental Protection. Invasive Plants of the Thirteen Southern States. http://www.invasive.org/seweeds.cfm

FLEPPC. 2003. Florida Exotic Pest Plant Council. List of invasive plants. http://www.fleppc.org/Plantlist/03list.htm

HALBERT, S. E. 2000. Entomology section. Trilogy 39(5) http://doacs.state.fl.us/~pi/enpp/00-sep-oct.htm#ent.

JOHNSON, S. R., AND A. K. KNAPP. 1996. Impact of *Ischnodemus falicus* (Hemiptera: Lygaeidae) on photosynthesis and production of *Spartina pectinata* wetlands. Environ. Entomol. 25: 1122-1127.

LANGELAND, K. A., AND K. CRADDOCK-BURKS. 1998. Identification and Biology of Non-native Plants in Florida's Natural Areas. University Press of Florida, Gainesville. 165 pp.

MEDINA, E., AND N. MOTTA. 1990. Metabolism and distribution of grasses in tropical flooded savannas in Venezuela. J. Tropical Ecology 6: 7-89.

MEYER, G. A., AND T. H. WHITLOW. 1992. Effects of leaf and sap feeding insects on photosynthetic rates of goldenrod. Oecologia 92: 480-489.

NEGRON, J. F., AND T. J. RILEY. 1990. Long-term effects of chinch bug (Hemiptera: Heteroptera: Lygaeidae) feeding on corn. J. Econ. Entomol. 83: 618-620.

PRESS, M. C., AND J. B. WHITTAKER. 1993. Exploitation of the xylem stream by parasitic organisms. Phil. Trans. Royal Soc., London B 341: 101-111.

SLATER, J. A. 1987. The taxonomic status of *Ischnodemus oblongus* (Fabricius) and *Ischnodemus variegatus* (Signoret) (Hemiptera: Lygaeidae: Blissinae). J. N.Y. Entomol. Soc. 95: 294-297.

TEJOS, M. R. 1980. Production of water straw grass (*Hymenachne amplexicaulis* (Rudge) Nees) during a savanna period. Congreso Venezolano Zootecnia Guanare (Venezuela) p. 24.

UNIVERSITY OF FLORIDA HERBARIUM. 2003. Collections catalogue. http://www.flmnh.ufl.edu/scripts/dbs/herbs_project/herbsproject/herbs_pub_proc.asp.

WALLING, L. L. 2000. The myriad of plant responses to herbivores. J. Plant Growth Regulation 19: 195-216.