

# Viability of Hydrilla Fragments Exposed to Different Levels of Insect Herbivory

CHETTA S. OWENS<sup>1</sup>, MICHAEL J. GRODOWITZ<sup>2</sup>, R. MICHAEL SMART<sup>3</sup>,  
NATHAN E. HARMS<sup>4</sup> AND JULIE M. NACHTRIEB<sup>4</sup>

## INTRODUCTION

Hydrilla (*Hydrilla verticillata* (L.f.) Royle) is an invasive, nonindigenous aquatic plant that was first discovered in the United States in the 1960s (Pieterse 1981). Current distribution of hydrilla in the United States includes the northern states of Maine and Washington, the Gulf and Atlantic coastal states, the western states of Arizona and California, Tennessee and recently Arkansas (USGS 2006). Once hydrilla invades an aquatic system, the plant can rapidly spread locally through rhizome expansion or over longer distances through generation of allofragments (hereafter called fragments) and/or turions. Unlike Eurasian watermilfoil (*Myriophyllum spicatum* L.) that forms autofragments for deliberate dispersal or due to nutrient depletion (Smith et al. 2002), hydrilla fragments can be generated by outside factors, such as recreational activities (Owens et al. 2001), wildlife, flooding events, boating traffic (Owens unpubl. data, Sculthrope 1985) or herbivory (Grodowitz unpubl. data).

Although there are no native insect herbivores that feed exclusively on hydrilla in the United States, two introduced host-specific leaf-mining flies have shown success at long-term management of hydrilla in controlled experimentation and field sites (Doyle et al. 2002, Grodowitz et al. 2003). The two introduced agents include the Australian leaf-mining fly (*Hydrellia balciunasi* Bock) and the Asian leaf-mining fly (*H. pakistanae* Deonier). The larval life stages (three-instars) damage the plant by penetrating, mining and destroying hydrilla leaves (Balciunas et al. 2002, Buckingham and Grodowitz 2004). Past research has shown that moderate to high levels of herbivory can impact hydrilla biomass production and reduce tuber numbers and size (Doyle et al. 2002, Doyle et al. 2006 (in review), Grodowitz et al. 2003, 2006). Doyle et al. (2002) reported that when 10-30% of leaves were damaged, the maximum rate of light-saturated photosynthesis was reduced by almost 40%. When leaf damage reached 70%, photosynthetic rates were reduced by up to 60%. This type of damage can impact hydrilla's ability to balance daily respiratory needs (Doyle et al. 2002, Grodowitz et al. 2003).

Based upon observations of field damaged hydrilla stems, fragmentation appears to be higher in hydrilla stems heavily damaged by fly mining (Grodowitz pers observ). If greater fragmentation occurs due to increased fly damage to hydrilla stems, how viable are these fragments? To answer this question, this initial study focused on how viable is hydrilla fragments under ideal conditions following leaf mining herbivory that resulted in low, medium, or high leaf damage.

## MATERIALS AND METHODS

The study was conducted at the U.S. Army Corps of Engineers Lewisville Aquatic Ecosystem Research Facility (LAERF) in Lewisville, TX over a four-week period during August 2005. Apical fragments measuring approximately 20 cm were collected from fly damaged hydrilla plants. The stems were separated into three damage categories based on low (0-30%), medium (40-60%), or high (70-100%) percent leaf damage. Thirty fragments from each damage level were planted one fragment per 1 L container in LAERF pond sediment (Smart et al. 1995) amended with 0.71 g ammonium sulfate and randomly placed in 60 cm tanks filled with alum-treated Lake Lewisville water.

After four weeks, hydrilla shoot biomass from each container was collected, measured for maximum height (cm) and total number of stems counted. The samples were dried to a constant weight. The container sediment was washed to extract tubers and rhizomes. Following counting, the belowground biomass samples were dried to a constant weight. One-way ANOVA's were performed on all parameters. Significant differences between means were determined using Tukey's level of significance. Statistics were performed using Statistix (Analytical Software, Tallahassee, FL).

## RESULTS AND DISCUSSION

High levels of herbivory significantly reduced all parameters measured including those for productivity (above and belowground biomass, stem length, and stem number) and vegetative reproduction (rhizome and tuber number) compared to low levels of herbivory (Figure 1A-F). Compared to medium levels of herbivory, high herbivory significantly reduced only those parameters that measured productivity (Figure 1 A-D). While no differences were found between low and medium damaged fragments for all parameters except aboveground biomass (Figure 1A), there was an almost three-fold difference in aboveground and belowground biomass between highly damaged fragments and those with leaf damage less than 30% (Figure 1A, B). It was obvious that fragments

<sup>1</sup>SpecPro-Lewisville Aquatic Ecosystem Research Facility, 201 Jones St., Lewisville, TX 75056. Corresponding author e-mail: chetta@laerf.org. Received for publication January 17, 2006 and in revised form March 21, 2006.

<sup>2</sup>U.S. Army Corps of Engineers Engineer Research and Development Center, 3909 Halls Ferry Rd., Vicksburg, MS 39180.

<sup>3</sup>USAE- Lewisville Aquatic Ecosystem Research Facility, 201 Jones St., Lewisville, TX 75056.

<sup>4</sup>University of North Texas- Lewisville Aquatic Ecosystem Research Facility, 201 Jones St., Lewisville, TX 75056.

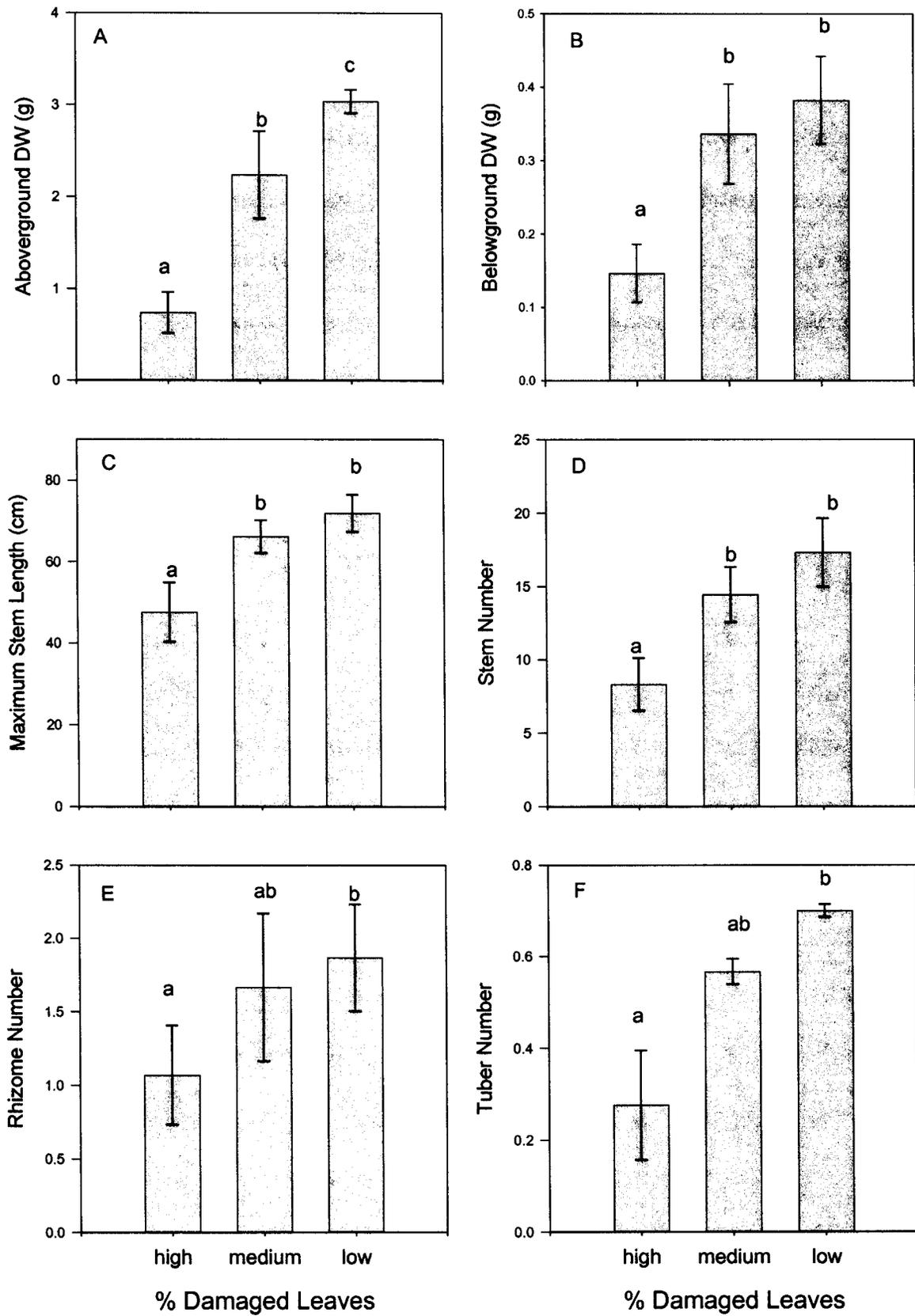


Figure 1. Comparison of hydrilla subjected to high, medium, and low percent herbivory damage: (A) aboveground biomass; (B) belowground biomass; (C) maximum stem length; (D) stem number; (E) rhizome number; and (F) average tuber number. Error bars represent 95% confidence interval. Means with the same letter are not significantly different at the  $p = 0.05$  level.

with greater than 70% leaf damage were significantly less viable than fragments with less herbivory. Not only were they structurally impaired but also the transparency of individual leaves indicated they were clearly unable to photosynthesize (Owens, pers observ). Hydrilla was apparently unable to overcome the effects of such a high level of herbivore damage and as a result there was a significant reduction in the ability to produce aboveground and belowground biomass. Low and medium damaged fragments were apparently able to outgrow herbivore effects and both above and belowground biomass significantly increased during the duration of the study. Similar results were observed for stem length and stem number with an almost two-fold increase in length and stem number between high and low damaged fragments (Figure 1C, D).

In addition to impacting the ability of hydrilla to produce biomass, high levels of herbivory negatively impacted the ability of hydrilla to produce tubers. Fragments with low leaf damage produced nearly three-times as many tubers compared to highly damaged fragments (Figure 1F). Remarkably, in this study, tubers were produced from fragments after only four weeks of growth. All tubers were found still attached to rhizomes for all treatments indicating that plants were still transporting reserves to the tubers. While significantly fewer tubers were produced from hydrilla exposed to high levels of herbivory (Figure 1F), the average dry weight of the tubers from the high herbivory containers was two-fold higher (0.08 g) compared to the average dry weight of tubers produced by low damaged fragments (0.03 g). These data could be misleading as so few tubers (8 total) were produced from the heavily damaged fragments compared to the low to medium damaged fragments where the tubers (21 total each treatment) were highly variable in size (0.01 to 0.07 g). Although the tubers produced from the low to medium damaged fragments were smaller overall, since all tubers were still attached to the stolons, nutrients and carbohydrates were still being transported to the attached tubers. The average dry weight of tubers collected at the LAERF in August 1995 generally ranged from 0.001-0.08 g however by October 1995 the dry weights had increased to 0.02-0.18 g as the tubers increased in size (Madsen and Owens 1998). Although the tubers were larger for the heavily damaged fragments, there were significantly less produced as compared to the numbers produced by the low to medium damaged fragments. If the experiment had continued, all attached tubers would have increased in size. These findings warrant further investigations.

It should also be noted that all fragments used in this study were planted and grown under conditions of a best-case scenario. Under natural field conditions, fragments could free-float for extended periods thus potentially lowering viability success of individual fragments especially after being heavily damaged from leaf mining. This study placed an artificially high level of success upon all study fragments. This is indicated by 90% stem fragment survival for the highly damaged stems with 100% survival observed for both the medium and low damaged stems. In a previous study focusing on establishment success of hydrilla fragments, Owens and Smart (In review) found that only 30 and 45% of free-floating undamaged hydrilla fragments settled when allowed to free-float in 5 L containers with less than 10% of the frag-

ments producing roots and anchoring in four weeks. In the current study, the fragments were deliberately planted thus affording greater establishment success. It would seem plausible that under free-floating conditions, lower survival would be noted for all fragments especially the highly damaged stems. Future studies need to address hydrilla fragments exposed to different levels of herbivory under free-floating conditions, allowing the fragments to naturally establish to provide more accurate information on hydrilla fragment viability under different levels of leaf-mining.

## ACKNOWLEDGMENTS

This research was conducted under the U.S. Army Corps of Engineers Aquatic Plant Control Research Program, U.S. Army Corps of Engineers Research and Development Center under the program leadership of Robert Gunkel. Permission to publish this information was granted by the Chief of Engineers. We would like to thank Dr. Judy Shearer and LeeAnn Glomski for review of this paper.

## LITERATURE CITED

- Balcianas, J. K., M. J. Grodowitz, A. F. Cofrancesco and J. F. Shearer. 2002. Hydrilla, pp. 91-114. *In: Biological Control of Invasive Plants in the Eastern United States*. R. Van Driesche, S. Lyon, B. Blossey, M. Hoddle, and R. Reardon (editors) USDA Forest Serv. Publ. FHTET-2002-04, Morgantown, WV.
- Buckingham, G. R. and M. J. Grodowitz. 2004. Hydrilla, pp. 184-195. *In: E. M. Coombs, J. K. Clark, G. L. Piper and A. F. Cofrancesco, Jr. (eds.). Biological control of invasive plants in the United States*. Oregon State University Press, Corvallis, OR.
- Doyle, R. D., M. J. Grodowitz, R. M. Smart and C. S. Owens. 2002. Impact of herbivory by *Hydrellia pakistanae* (Diptera: Ephydriadae) on growth and photosynthetic potential of *Hydrilla verticillata*. *Biol. Control* 24:221-229.
- Doyle, R.D., M.J. Grodowitz, R. M. Smart and C.S. Owens. 2006. Separate and interactive effects of competition and herbivory on the growth, expansion, and tuber formation of *Hydrilla verticillata*. *Biol. Control* (In review).
- Grodowitz, M. J., R. M. Smart, R. D. Doyle, C. S. Owens, R. Bare, C. Snell, J. Freedman and H. Jones. 2003. *Hydrellia pakistanae* and *H. balciunasi* insect biological agents of hydrilla: boon or bust? pp. 529-538. *In: J. M. Cullen, D. T. Briese, D. J. Kriticos, W. M. Lonsdale, L. Morin and J. K. Scott (eds.). Proceedings of the XI International Symposium on Biological Control of Weeds*, Canberra, Australia.
- Grodowitz, M. J., C. S. Owens, R. M. Smart and J. M. Graham. 2006. Impact of Herbivory and Plant Competition on the Growth of Hydrilla in Small Ponds. Corps of Engineers' Technical Report (In review).
- Madsen, J. D. and C. S. Owens. 1998. Seasonal biomass and carbohydrate allocation in dioecious hydrilla. *J. Aquat. Plant Manage.* 36:138-145.
- Owens, C. S., J. D. Madsen, R. M. Smart and R. M. Stewart. 2001. Dispersal of native and nonnative aquatic plant species in the San Marcos River, Texas. *J. Aquat. Plant Manage.* 39:75-79.
- Owens, C. S. and R. M. Smart. 2006. Fragment viability and establishment of *Hydrilla verticillata* (L.f.) Royle. Corps of Engineers' Technical Report (In review).
- Pieterse, A. H. 1981. *Hydrilla verticillata*—a review. *Abstr. Trop. Agric.* 7(6):9-34.
- Sculthorpe, C. D. 1985. *The biology of aquatic vascular plants*. Edward Arnold (Publ.) Ltd., London, England. 610 pp.
- Smart, R. M., J. D. Madsen, J. R. Snow, G. O. Dick and D. R. Honnell. 1995. Physical and environmental characteristics of experimental ponds at the Lewisville Aquatic Ecosystem Research Facility. Technical Report A-95-2, U.S. Army Engineer Waterways Experiment Station, Vicksburg, MS. 22 pp.
- Smith, D. H., J. D. Madsen, K. L. Dickson and T. L. Beiting. 2002. Nutrient effects on autofragmentation of *Myriophyllum spicatum*. *Aquatic Bot.* 74:1-17.
- USGS. 2006. [http://nas.er.usgs.gov/taxgroup/plants/docs/hy\\_verti.html](http://nas.er.usgs.gov/taxgroup/plants/docs/hy_verti.html), USGS-NAS-Nonindigenous Aquatic Species, *Hydrilla verticillata* (L.f.) Royle, March 2006.









