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# Effects of damage and pollination on sexual and asexual reproduction in a flowering clonal plant

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**Abstract** The extent to which local biotic factors like herbivory and pollination affect mode of reproduction in plants is not yet fully understood. Mode of reproduction is ecologically important because it can influence the spread and distribution of plant populations through factors like offspring dispersal distance and establishment success. The two experiments described here address the potential effects of damage and pollen receipt on plant growth, sexual reproduction, and asexual reproduction in a clonal flowering plant (*Eichhornia crassipes*, water hyacinth). These experiments were conducted in greenhouse and outdoor tanks at the Florida State University research facilities in Tallahassee, FL, on plants collected from north Florida populations. Plants received manual damage to leaves (imposing a loss of resources), apical meristems (imposing a loss of leaf and flower production), or axillary meristems (imposing a loss of clone production). Apical meristem damage increased asexual reproduction (clone number). When severe, axillary meristem damage increased plant growth (leaf production). Neither leaf damage nor pollination affected plant growth, clone production, or flower production. Asexual reproductive responses to damage have not been well studied, although sexual reproduction and individual plant growth have been shown to increase following damage. These results have implications for the dispersal and establishment of clonal plants in the presence of herbivory. For a highly invasive species like *E. crassipes*, these results can further inform the use of insect herbivores to manage invasive populations.

**Keywords** Clonal plants · Herbivory · Mode of reproduction · Plant–insect interactions · *Eichhornia crassipes*

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## Introduction

Many plants are capable of reproducing both sexually via flowers and asexually via clone production. These two modes of reproduction have consequences for the distribution and abundance of individuals, for example by influencing offspring dispersal distance (Eriksson

1997) or rate of offspring establishment and survival (Herrera and Nassar 2009). Understanding how the local environment affects mode of reproduction allows us to more fully understand the spread of plant populations. Herbivores and pollinators are two important facets of a plant's local environment, and yet there are still gaps in our knowledge of how herbivores and pollinators influence mode of reproduction in clonal flowering plants.

Herbivores generally negatively affect traits that influence the ability of plants to reproduce sexually. Herbivory has been shown to reduce pollen production and performance (Quesada et al. 1995), reduce flower size (Lehtilä and Strauss 1997; Steets and Ashman 2004), and reduce the number of open flowers on a plant (Elle and Hare 2002). There is much less research on the relationship between herbivory and asexual reproduction, and even fewer studies measuring actual clone production in response to damage (but see Brathen and Junttila 2006). Some studies use a proxy like rhizome size to estimate asexual reproduction (e.g., Meyer and Root 1993; Wise et al. 2006), which in some cases may accurately reflect clonal offspring production, but in other cases does not (Boose and Holt 1999; Bai et al. 2009).

The effects of pollinators on plant sexual reproductive traits have been well studied. Pollen receipt can decrease the number of open flowers (Harder and Johnson 2005) and decrease flower longevity (Clark and Husband 2007). Failure to receive pollen can increase nectar volume and pollen receipt in subsequent flowers (Ladio and Aizen 1999) and can cause a shift from semelparity to iteroparity (Paige and Whitham 1987a). Although it is less likely that pollination could directly influence asexual reproduction in the same manner it influences sexual reproduction, changes in plant traits following pollination could indirectly influence clone production through costs of fruit production (e.g., Snow and Whigham 1989). There is still work to be done in understanding potential effects of pollination and herbivory particularly on plant asexual reproductive traits.

The two experiments described here address this gap, investigating how damage and pollination influence investment in leaf number (plant growth), flower number (sexual reproduction), and clonal offspring number (asexual reproduction) in the aquatic plant *Eichhornia crassipes* (water hyacinth).

## Materials and methods

### Study system

*Eichhornia crassipes* (Mart.) Solms (Commelinales: Pontederiaceae) is a freshwater aquatic perennial plant, native to South America and invasive in many regions of the world (Penfound and Earle 1948). All leaves, roots, and inflorescences radiate from a central rhizome (Penfound and Earle 1948). An apical meristem produces leaves until inflorescence production begins. There is an additional, axillary meristem associated with each new leaf. Unlike axillary meristems of many plants, which produce new branches on the original plant, axillary meristems in *E. crassipes* can either produce clonal daughter plants or continuation stems capable of making more leaves or an additional inflorescence (Geber et al. 1992). Inflorescences produce many lavender flowers (in this study, mean flower number per inflorescence = 6.4, SD = 2.2, $n = 117$), which in their native range are typically pollinated by specialist bees (Zhang et al. 2010) or other insect visitors (Barrett 1980). Flowers are self-compatible and some invasive populations produce seeds despite herkogamy and a lack of pollinators (Penfound and Earle 1948). However, seed germination is rare in many invasive populations, making asexual reproduction the primary means of population growth in invaded ranges (Zhang et al. 2010).

### Experiment 1: damage only (2010)

*Eichhornia crassipes* individuals ($n = 120$) were collected from three populations in north Florida (Fig. 1): the Crooked River (N29.92872°, W84.62568°; hereafter "CRK"), Lake Munson (N30.37000°, W84.31465°; hereafter "MUN"), and the Wacissa River (N30.34384°, W83.99923°; hereafter "WAC"). Plants were collected from different populations to increase genetic diversity but due to differences in population size, populations were not equally represented ($n = 38$ CRK, $n = 8$ MUN, $n = 74$ WAC). After collection, plants were transported to Florida State University Mission Road Research Facility in Tallahassee, FL, where they were immediately inspected and cleared of herbivores. Each of the 120 field-collected plants received one of four damage treatments and were distributed among six outdoor cattle tanks filled with

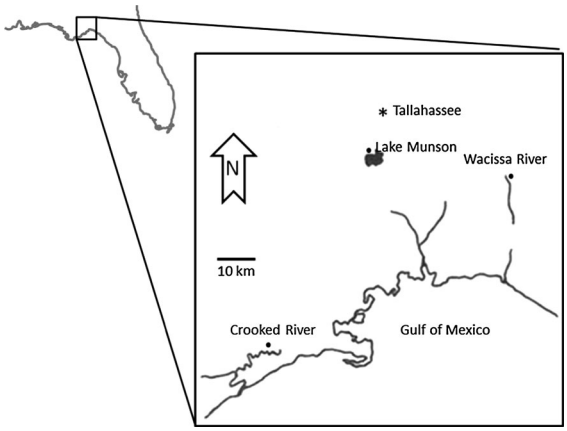


**Fig. 1** Map of the three north Florida *Eichhornia crassipes* populations sampled for these experiments. Survey and collection sites are marked with a *solid circle*. Tank experiments took place at the Mission Road greenhouse facility in Tallahassee, FL, marked with an *asterisk*

325 L of well water. Plants were distributed as evenly as possible given the population sample sizes (20 plants per tank, five from each treatment, with plants from each population in each tank).

This experiment consisted of three damage treatments and one undamaged control treatment ($n = 30$ per treatment). Leaf damage was imposed with a razor blade, removing approximately 25–30 % of the plant's leaf surface tissue; this level of damage was greater than that observed in natural populations (Electronic Supplementary Materials, Table A1). Apical and axillary damage treatments were imposed with sharp forceps, destroying all meristem tissue down to the rhizome, which is identifiable by its pink cap (Penfound and Earle 1948). The amount of meristem damage was not measured in natural populations, but destruction of all apical or axillary meristem tissue is likely more severe than natural meristem damage levels. Control plants were not damaged, but were handled similarly to damaged plants to account for handling effects. Damage types were intended to directly influence three important aspects of plant function: leaf damage directly reduces photosynthetic leaf area, apical damage directly prevents leaf and inflorescence production, and axillary damage directly prevents clone production. These damage treatments, while artificial, do target plant parts commonly damaged by water hyacinth weevils *Neochetina eichhorniae* and *N. bruchi*, important *E. crassipes* herbivores. Leaf damage was distributed evenly in small patches across expanded leaves, mimicking natural adult herbivore damage patterns (Electronic Supplementary Materials, Fig. A1); larval herbivores damage meristems as well as other internal plant parts (DeLoach and Cordo 1976; Stark and Goyer 1983; Cilliers 1991; Center et al. 2005). Measuring subsequent flower, clone, and leaf production will demonstrate the direct effect of damage on the damaged plant part (i.e., whether the treatments were effective) and the indirect effect on other plant functions.

Sexual and asexual reproduction and plant growth were measured at 37 days post-damage (DPD); due to the rapid growth rate of *E. crassipes*, this was more than enough time for experimental plants to produce inflorescences and several clones. Flower production was recorded as a binary response, whether or not each plant produced an inflorescence during the experiment. Clone production was recorded as the total number of new clones produced between the beginning and end of the experiment. Leaf production was recorded as the difference in leaf number between the beginning and end of the experiment.

Separate analyses of covariance (ANCOVA, Type III SS) were performed on leaf and clone production. Initial models included damage treatment, initial leaf number (because initial leaf number was part of the calculation for leaf production response, this variable was included in clone response analysis only), initial plant mass, source population, and tank. For this and all other analyses, tank and population were included to account for variance rather than to generalize about population differences or interpreting effects of tank identity, and as such were treated as fixed effects blocking factors (Bates 2010). Model selection in this and all other analyses used hierarchical elimination of non-significant interactions, where applicable, and non-significant covariates. Final models are presented in Table 1. For this and all other analyses, where ANCOVAs yielded significant main effects, Tukey's HSD determined differences between treatments within that effect. All analyses were performed in R 2.13.2 (R Development Core Team 2011).

## Experiment 2: damage and pollination (2011)

*Eichhornia crassipes* individuals ($n = 304$) were collected from two source populations ($n = 152$ WAC, $n = 152$ CRK). After collection, plants were transported to Florida State University Mission Road

**Table 1** ANOVA (Type III SS) table reporting responses to damage treatments in 2010 and 2011

| Response | | Predictor | *Df* | *F* | *P* |
|---|---|---|---|---|---|
| Damage-only experiment | Change in leaf number, 37 DPD | Damage treatment | 3 | 12.82 | <0.001 |
| | | Source population | 2 | 8.2 | <0.001 |
| | | Tank | 5 | 2.41 | <0.05 |
| | New clones, 37 DPD | Residuals | 109 | | |
| | | Damage treatment | 3 | 3.62 | <0.05 |
| | | Initial leaf number | 1 | 8.9 | <0.01 |
| | | Plant mass | 1 | 13.43 | <0.001 |
| | | Residuals | 114 | | |
| Damage & pollination experiment | Change in leaf number, 11 DPD | Damage treatment | 3 | 113.29 | <0.001 |
| | | Plant mass | 1 | 17.79 | <0.001 |
| | | Tank | 37 | 2.1 | <0.001 |
| | | Residuals | 262 | | |
| | Change in leaf number, 27 DPD | Damage treatment | 3 | 182.42 | <0.001 |
| | | Plant mass | 1 | 7.26 | <0.01 |
| | | Tank | 37 | 4.76 | <0.001 |
| | | Residuals | 261 | | |
| | New clones, 11 DPD | Damage treatment | 3 | 314.12 | <0.001 |
| | | Source population | 1 | 8.74 | <0.01 |
| | | Residuals | 299 | | |
| | New clones, 27 DPD | Damage treatment | 3 | 11.49 | <0.001 |
| | | Initial leaf number | 1 | 22.46 | <0.001 |
| | | Residuals | 298 | | |

*DPD* days post-damage

Research Facility in Tallahassee, FL, where they were immediately inspected and cleared of herbivores. The 304 field-collected plants were distributed among 38 indoor greenhouse glass tanks filled with 50 L of well water. Each tank held four plants from each population, each of which was assigned one of four damage treatments. Tanks were filled with well water and fertilized as needed with 2–4 g/L of Suncote 15-8-11 slow release fertilizer and 0.2 g/L Miller Iron Chelate DP 10 % Fe.

In this experiment, plants received both damage and pollination. Damage treatments were identical to those in experiment 1 ($n = 76$ in each damage treatment), except that axillary damage was continuous over 10 days and resulted in more meristems being damaged. Apical and leaf damage were imposed on the final day of axillary damage, so for all damage types in this experiment, days post-damage (DPD) refers to the number of days after all damage was complete. As plants began to develop flowers (4–11 DPD), they were assigned to receive either hand pollination ($n = 31$) where all flowers on a plant received pollen, or no pollination ($n = 33$) where no flowers received pollen. Pollinated flowers received a mix of pollen from at least five plants from a different source population, in order to reduce the likelihood of receiving pollen from a genetically identical individual. Pollination and damage treatments were crossed, so that each damage treatment group (except for apically damaged plants, which did not produce flowers) had approximately equal numbers of pollinated and non-pollinated plants.

Due to the rapid growth rate of *E. crassipes*, responses in this experiment were measured at 11 DPD (responses to damage) and 27 DPD (responses to both damage and pollination, and interactive effects of damage and pollination). Rapid plant growth also necessitated transferring all plants to outdoor cattle tanks at 14 DPD, combining 18–20 plants from multiple indoor tanks per outdoor tank and keeping approximately equal numbers of plants from each treatment in each tank. Number of surviving plants

was recorded at 27, 41, 60, 80, and 100 DPD, at which time the experiment ended.

Because not all plants flowered, main effects of damage ($n = 304$) were analyzed separately from the main effects of pollination and the interactive effects of damage and pollination ($n = 64$). The effects of damage on leaf production at 11 and 27 DPD were analyzed with separate ANCOVAs, where initial models included damage treatment, initial plant mass, source population, and original tank as predictor variables. As in experiment 1, tank and population were included to account for variance rather than to generalize about population differences or interpreting effects of tank identity, and as such were treated as fixed effects blocking factors (Bates 2010). The effects of damage on clone production at 11 and 27 DPD were analyzed with a generalized least squares model using the R package "nlme" (Pinheiro et al. 2011). The generalized least squares model was used to account for heterogeneity of variance among damage treatments (Zuur et al. 2009). Initial models included damage treatment, initial leaf number, initial plant mass, source population, and original tank. Results from separate analyses on responses at 11 and 27 DPD will be reported, rather than a single repeated measures analysis, to facilitate comparisons with the 37 DPD responses in experiment 1 and with the 27 DPD responses to both damage and pollination in this experiment, described below. Number of surviving individuals and flower presence were each analyzed using Chi squared tests with damage treatment as the lone predictor. Fruit number per pollinated flower and seed mass per fruit were analyzed with ANCOVAs, with damage treatment and source population as predictors.

To quantify main effects of pollination and interactive effects of damage and pollination, clone and leaf production at 27 DPD were analyzed with ANCOVAs. Initial models included pollination treatment, damage treatment, initial leaf number (for clone response only), initial mass, original tank, and a damage-by-pollination interaction.

## Results

### Experiment 1: damage only (2010)

Damage treatment affected both leaf and clone production at 37 DPD (Table 1; Fig. 2). The apical damage treatment was effective, in that plants with apical damage lost leaves compared with control plants (Table 2; Fig. 2). Plants with apical damage also produced 44 % more new clones compared with controls (Table 2; Fig. 2). However, the axillary damage treatment was not effective in this experiment (clone production was not reduced), so it is not surprising that there was no effect on leaf production (Fig. 2). Leaf damage affected neither leaf nor clone production (Fig. 2).

### Experiment 2: damage and pollination (2011)

Damage treatments affected leaf and clone production 11 and 27 DPD (Table 1; Fig. 3). The apical damage treatment was effective in this experiment: plants with apical damage lost leaves compared with control plants (Table 2; Fig. 3a, b). In contrast to the 2010 experiment, axillary damage was effective in the 2011 experiment: plants with axillary damage produced at least 50 % fewer new clones than control plants 11 and 27 DPD (Table 2; Fig. 3c, d). In addition, plants with axillary damage produced 40 % more leaves compared with control plants 11 DPD (Table 2; Fig. 3a), although this effect disappeared by 27 DPD (Fig. 3b). For reasons addressed in the methods, separate ANOVAs were used to analyze each subset of data for this experiment; however, repeated measures analysis of leaf and clone responses to damage treatments only yielded qualitatively comparable results (leaf number $F_{3,597} = 3.095$, $P = 0.0265$; clone number $F_{3,597} = 6.422$, $P = 0.0003$).

This experiment showed, not surprisingly, that plants with apical damage were 70 % less likely to flower over the entire 100 day experiment ($\chi^2 = 24.23$, $n = 304$, $df = 3$, $P < 0.0001$; Fig. 4). Plants with apical damage were also 60 % more likely than any other damage group to die before the end of the experiment ($\chi^2 = 32.68$, $n = 304$, $df = 12$, $P = 0.001$; Fig. 5). As in experiment 1, leaf damage did not affect leaf or clone production (Table 1; Fig. 3).

For those plants that were part of the pollination treatments, there was no significant interaction between damage and pollination ($P > 0.05$), nor were there any main effects of pollination treatment on leaf or clone production ($P > 0.05$). Plants receiving hand pollination set fruit (90.3 % fruit set rate, compared to 0 % fruit set in non-pollinated plants) and produced seeds, but none of the damage treatments influenced fruit set or seed mass ($P > 0.05$).

**Table 2** *P*-values for Tukey's HSD post hoc comparisons among damage groups for leaf and clone production responses. *P* values in bold are significant to < 0.05

| | | | Control | Apical | Axillary |
|---|---|---|---|---|---|
| Damage-only experiment | Change in leaf number, 37 DPD | Apical | **<0.0001** | | |
| | | Axillary | 0.84 | **<0.001** | |
| | | Leaf | 0.81 | **<0.001** | 0.99 |
| | New clones, 37 DPD | Apical | **0.04** | | |
| | | Axillary | 0.99 | 0.06 | |
| | | Leaf | 0.99 | **0.02** | 0.97 |
| Damage & pollination experiment | Change in leaf number, 11 DPD | Apical | **< 0.0001** | | |
| | | Axillary | **0.04** | **<0.0001** | |
| | | Leaf | 0.93 | **<0.0001** | 0.16 |
| | Change in leaf number, 27 DPD | Apical | **<0.0001** | | |
| | | Axillary | 0.84 | **<0.0001** | |
| | | Leaf | 0.85 | **<0.0001** | 0.99 |
| | New clones, 11 DPD | Apical | 0.51 | | |
| | | Axillary | **< 0.0001** | **< 0.001** | |
| | | Leaf | 0.99 | 0.67 | **<0.0001** |
| | New clones, 27 DPD | Apical | 0.06 | | |
| | | Axillary | **<0.0001** | **0.003** | |
| | | Leaf | 0.80 | 0.36 | **<0.0001** |

*DPD* days post-damage

## Discussion

Despite considerable evidence for how herbivore damage and pollination can influence plant sexual reproductive traits, much less is known about the effects of damage and pollination on asexual reproductive traits. Results from this study suggest that in some cases damage to structures involved in one mode of reproduction can influence investment in another mode of reproduction. Apical meristem damage influenced asexual reproduction in one experiment, and axillary meristem damage influenced plant growth in another experiment. Research in other plant systems has shown that damage to apical meristems can result in the production of more flowers or lateral branches (Paige and Whitham 1987b; Pilson and Decker 2002), but a similar response in clone production has not been demonstrated. Likewise, studies on plant responses to damaged asexual reproductive structures, such as axillary meristems, are exceedingly rare. Previous research has found that high-density herbivore infestations can lead to increased leaf production in *E. crassipes* (Center and Van 1989), but because these herbivores infested entire plants, the specific cause of increased leaf production is not clear.

The plant responses observed in this experiment may help *E. crassipes* populations persist in the face of heavy herbivory. Producing more clones after apical damage might help ensure reproduction before the parent plant dies, as individual mortality following apical meristem damage is high, possibly due to the loss of plant buoyancy that new leaves provide (Penfound and Earle 1948). Producing more leaves after axillary damage might indirectly compensate for lost axillary meristems and enable plants to maintain asexual reproduction, because leaf production enables axillary meristem production (Geber et al. 1992).

There is an additional temporal trend across the two experiments that the experimental design was not capable of fully resolving. Although apical damage was imposed identically in both experiments, apical damage increased clone production only in the longer-term survey (experiment 1), but not in the shorter-term surveys (experiment 2). Although the two experiments are too different to compare directly, a reasonable conclusion is that clonal responses to apical damage may take several weeks to appear.

There is also the suggestion of a threshold response across the two experiments, again which could not be

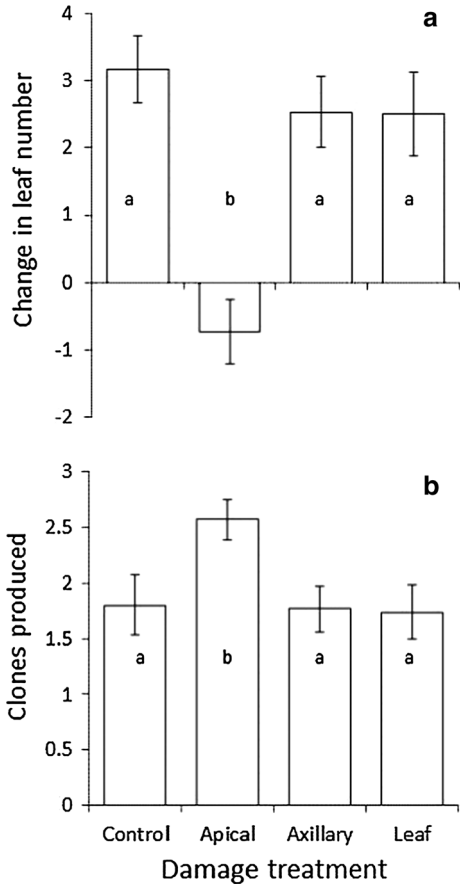


**Fig. 2** Plant responses to damage treatments in experiment 1 (damage only). Mean change in leaf number (**a**) and mean number of new clones produced (**b**) 37 days post-damage. Bars encoded with different lower-case letters were significantly different from each other at $P < 0.05$, using Tukey's HSD. *Error bars* show mean ± SE, $n = 119$

resolved given the experimental design. Axillary damage only influenced leaf production in experiment 2, where axillary damage was imposed over 10 days as opposed to a single damage event in experiment 1. In addition, leaf responses to axillary damage were only evident in the early surveys in experiment 2 but not in later surveys in either experiment, suggesting an immediate but not lasting effect of axillary damage.

In contrast to meristem damage, leaf damage did not affect any measured response in either experiment. This was despite the fact that experimental damage levels were about threefold more severe than the maximum leaf damage observed in the natural populations (Electronic Supplementary Materials, Table A1). Because meristem tissues produce hormones with wide-ranging effects on plant growth, and meristem damage can cause large plant responses such as lateral branching (Paige and Whitham 1987b), it is not entirely surprising that meristem damage had the most profound effect on *E. crassipes* in these experiments. Overall, this experiment demonstrates that meristem damage in *E. crassipes* can influence plant structures not directly damaged. Further, the influence of meristem damage on leaf and clone production, and a lack of any influence on flower production, indicate that *E. crassipes* shows a stronger and overall positive response of growth and asexual reproduction to meristem damage, relative to sexual reproduction.

The damage imposed on leaves and meristems in these experiments is typical of the type of damage done by the specialist herbivores *N. eichhorniae* (mottled water hyacinth weevil) and *N. bruchi* (chevroned water hyacinth weevil), which feed on *E. crassipes* as adults and larvae. Because *Neochetina* spp. larvae tunnel through plant tissues (DeLoach and Cordo 1976; Stark and Goyer 1983; Cilliers 1991) and damage meristems (Center et al. 2005), they may be more likely to affect *E. crassipes* than are *Neochetina* spp. adults, which only feed on the leaf surface. While the damage imposed in these experiments mimics *Neochetina* spp. adult and larval damage, these results are likely a conservative estimate of the effects of actual damage in field populations, and may underestimate these effects. Separate experiments showed that damage by adult *Neochetina* spp. weevils induced systemic defenses in *E. crassipes*, while damage mimicking adult and larval herbivory did not (Buchanan 2013). It is not yet known whether real larval damage induces resistance, but internal feeders can induce resistance in other plant species (Diezel et al. 2011; Ralph et al. 2006). Because the study described here used artificial damage, it is unlikely that plant defenses were induced, unlike plant responses to real herbivory. If production of induced defenses following insect herbivory is costly, the results reported here might underestimate the effects of herbivore damage on reproduction.

The results reported here also draw attention to the effects of herbivory on invasive plant populations. Plants invading a new range face a novel biotic environment that might influence their reproductive mode, and therefore their dispersal and establishment abilities. For the highly invasive *E. crassipes*, *Neochetina* spp. weevils have been shown to reduce plant

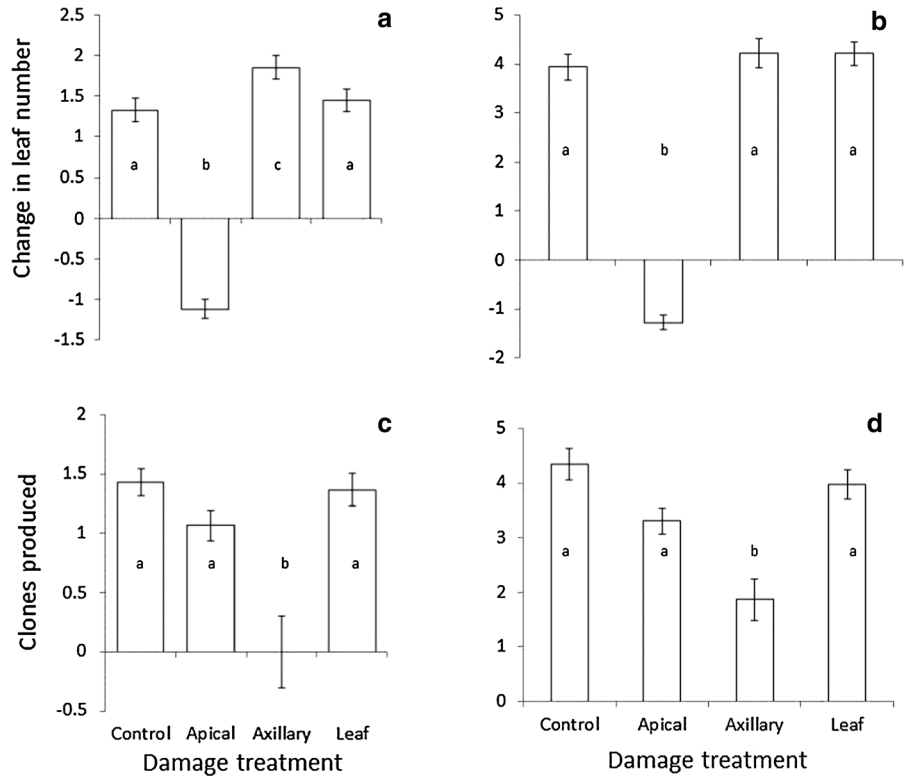


**Fig. 3** Plant responses to damage treatments in experiment 2 (damage & pollination). Mean change in leaf number 11 days (**a**) and 27 days (**b**) post-damage, and mean number of new clones 11 days (**c**) and 27 days (**d**) post-damage. *Bars* encoded with different lower-case letters were significantly different from each other at $P < 0.05$, using Tukey's HSD. *Error bars* show mean ± SE, $n = 302$ or 303

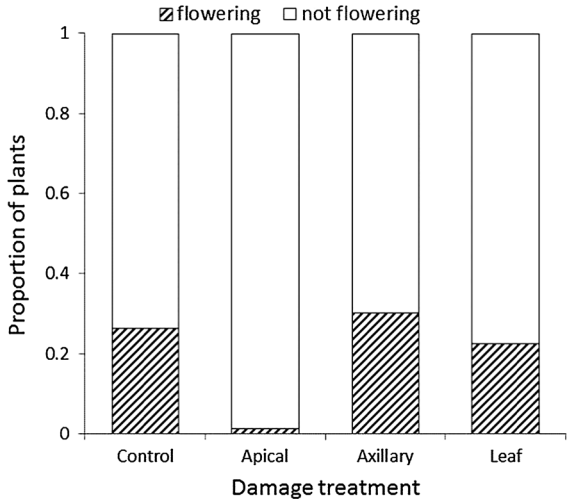


**Fig. 4** Influence of damage treatment on likelihood of flowering, damage, & pollination experiment ($\chi^2 = 24.23$, $n = 304$, $df = 3$, $P < 0.0001$)

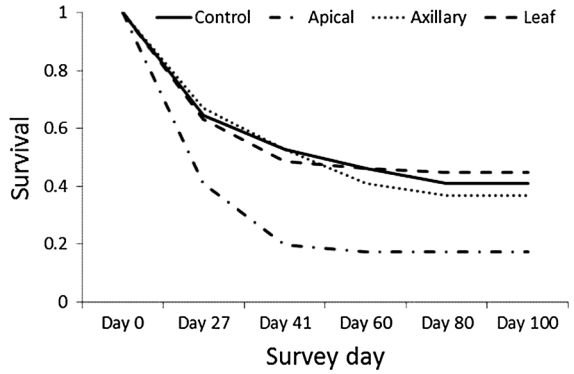


**Fig. 5** Proportion of individuals surviving in each damage treatment group, damage, & pollination experiment ($\chi^2 = 32.68$, $df = 12$, $n = 304$, $P = 0.001$)

growth (Center et al. 1999b). However, the experiments described here found an increase in clone production following damage, suggesting that *Neochetina* spp. biocontrol might be less effective when used as a sole management technique, and additional control methods like herbicide (Center et al. 1999a) or pathogens (Moran 2005) might be needed.

In this study, successful pollination did not influence any measured plant trait, nor was there an interactive effect of pollination and damage, even though all plants receiving pollen produced multiple fruits. While fruit production has been shown to

impose costs on growth or future reproduction in other systems (Harder and Johnson 2005; Obeso 2002 and references within), no such trade-offs were found here. In many invasive *E. crassipes* populations, failure to produce fruit does not appear to hinder population growth, which primarily occurs asexually (Zhang et al. 2010; Bock 1969). These high rates of clonal reproduction, coupled with low rates of pollination and seed germination, lead one to ask why invasive *E. crassipes* populations continue to invest in flower production. A contributing factor may be extremely low genetic variation within and among invasive populations (Zhang et al. 2010), prohibiting a response to potential selection for decreased sexual reproduction.

Overall, these experiments emphasize the need to explicitly measure both sexual and asexual reproductive responses to damage. Despite the ubiquity of clonal proliferation in plants, we still know little about clonal responses to a plant's biotic environment. Asexual reproductive responses could represent a major avenue of herbivore influence on plant populations that is not yet well understood.

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## References

Bai W, Sun X, Wang Z, Li L (2009) Nitrogen addition and rhizome severing modify clonal growth and reproductive modes of *Leymus chinensis* population. Plant Ecol 205:13–21

Barrett S (1980) Sexual reproduction in *Eichhornia crassipes* (water hyacinth). 2. Seed production in natural populations. J Appl Ecol 17:113–124

Bates, DM (2010) lme4: Mixed-effect modeling with R. http://www.lme4.r-forge.r-project.org/book/

Bock JH (1969) Productivity of water hyacinth *Eichhornia crassipes* (Mart) Solms. Ecology 50:460–464

Boose A, Holt J (1999) Environmental effects on asexual reproduction in *Arundo donax*. Weed Res 39:117–127

Brathen KA, Junttila O (2006) Infertile times: response to damage in genets of the clonal sedge *Carex bigelowii*. Plant Ecol 187:83–95

Buchanan A (2013) Damage by *Neochetina* weevils (Coleoptera: Curculionidae) induces resistance in *Eichhornia crassipes* (Commelinales: Pontederiaceae). Fla Entomol 96(2):458–462

Center T, Van T (1989) Alteration of water hyacinth (*Eichhornia crassipes* (Mart) Solms) leaf dynamics and phytochemistry by insect damage and plant density. Aquat Bot 35:181–195

Center T, Dray F, Jubinsky G, Grodowitz M (1999a) Biological control of water hyacinth under conditions of maintenance management: can herbicides and insects be integrated? Environ Manag 23:241–256

Center T, Dray F, Jubinsky G, Leslie A (1999b) Water hyacinth weevils (*Neochetina eichhorniae* and *N. bruchi*) inhibit water hyacinth (*Eichhornia crassipes*) colony development. Biol Control 15:39–50

Center T, Van T, Dray F, Franks S, Rebelo M, Pratt P, Rayamajhi M (2005) Herbivory alters competitive interactions between two invasive aquatic plants. Biol Control 33:173–185

Cilliers C (1991) Biological control of water hyacinth, *Eichhornia crassipes* (Pontederiaceae), in South-Africa. Agric Ecosyst Environ 37:207–217

Clark MJ, Husband BC (2007) Plasticity and timing of flower closure in response to pollination in *Chamerion angustifolium* (Onagraceae). Int J Plant Sci 168:619–625

DeLoach C, Cordo H (1976) Life-cycle and biology of *Neochetina bruchi*, (Coleoptera-Curculionidae-Bagoini) a weevil attacking waterhyacinth in Argentina, with notes on *Neochetina eichhorniae*. Ann Entomol Soc Am 69:643–652

Diezel C, Kessler D, Baldwin IT (2011) Pithy protection: *nicotiana attenuata*'s jasmonic acid-mediated defenses are required to resist stem-boring weevil larvae. Plant Physiol 155:1936–1946

Elle E, Hare JD (2002) Environmentally induced variation in floral traits affects the mating system in *Datura wrightii*. Funct Ecol 16:79–88

Eriksson O (1997) Clonal life histories and the evolution of seed recruitment. de Kroon H and Van Groenendael JM (eds) The ecology and evolution of clonal plants. Backhhuys Publishers, Leiden, pp 211–226

Geber M, Watson M, Furnish R (1992) Genetic differences in clonal demography in *Eichhornia crassipes*. J Ecol 80:329–341

Harder L, Johnson S (2005) Adaptive plasticity of floral display size in animal-pollinated plants. Proc R Soc Lond Ser B-Biol Sci 272:2651–2657

Herrera I, Nassar JM (2009) Reproductive and recruitment traits as indicators of the invasive potential of *Kalanchoe daigremontiana* (Crassulaceae) and *Stapelia gigantea* (Apocynaceae) in a Neotropical arid zone. J Arid Environ 73:978–986

Ladio AH, Aizen MA (1999) Early reproductive failure increases nectar production and pollination success of late flowers in south Andean *Alstroemeria aurea*. Oecologia 120:235–241

Lehtilä K, Strauss SY (1997) Leaf damage by herbivores affects attractiveness to pollinators in wild radish, *Raphanus raphanistrum*. Oecologia 111:396–403

Meyer G, Root R (1993) Effects of herbivorous insects and soil fertility on reproduction of goldenrod. Ecology 74:1117–1128

Moran P (2005) Leaf scarring by the weevils *Neochetina eichhorniae* and *N. bruchi* enhances infection by the fungus

*Cercospora piaropi* on water hyacinth *Eichhornia crassipes*. Biocontrol 50:511–524

Obeso JR (2002) The costs of reproduction in plants. New Phytol 155:321–348

Paige KN, Whitham TG (1987a) Flexible life history traits: shifts by scarlet gilia in response to pollinator abundance. Ecology 68(6):1691–1695

Paige KN, Whitham TG (1987b) Overcompensation in response to mammalian herbivory: the 340 advantage of being eaten. Am Nat 129:407–416

Penfound W, Earle T (1948) The biology of the water hyacinth. Ecol Monogr 18:447–472

Pilson D, Decker K (2002) Compensation for herbivory in wild sunflower: response to simulated damage by the head-clipping weevil. Ecology 83:3097–3107

Pinheiro J, Bates D, DebRoy S, Sarkar D (2011) Nlme: linear and nonlinear mixed effects models, R package version 3.1-102

Quesada M, Bollman K, Stephenson AG (1995) Leaf damage decreases pollen production and hinders pollen performance in *Cucurbita texana*. Ecology 76:437–443

Ralph SG, Yueh H, Friedmann M, Aeschliman D, Zeznik JA, Nelson CC, Butterfield YSN, Kirkpatrick R, Liu J, Jones SJM, Marra MA, Douglas CJ, Ritland K, Bohlmann J (2006) Conifer defence against insects: microarray gene expression profiling of Sitka spruce (*Picea sitchensis*) induced by mechanical wounding or feeding by spruce budworms (*Choristoneura occidentalis*) or white pine weevils (*Pissodes strobi*) reveals large-scale changes of the host transcriptome. Plant Cell Environ 29:1545–1570

Snow A, Whigham D (1989) Costs of flower and fruit production in *Tipularia discolor* (Orchidaceae). Ecology 70:1286–1293

Stark J, Goyer R (1983) Life-cycle and behavior of *Neochetina eichhorniae* Warner (Coleoptera, Curculionidae) in Louisiana: a biological-control agent of waterhyacinth. Environ Entomol 12:147–150

Steets JA, Ashman TL (2004) Herbivory alters the expression of a mixed-mating system. Am J Bot 91:1046–1051

R Development Core Team 2011 R: a language and environment for statistical computing

Wise MJ, Abrahamson WG, Landis K (2006) Edaphic environment, gall midges, and goldenrod clonal expansion in a mid-successional old-field. Acta Oecol-Int J Ecol 30:365–373

Zhang Y, Zhang D, Barrett SCH (2010) Genetic uniformity characterizes the invasive spread of water hyacinth (*Eichhornia crassipes*), a clonal aquatic plant. Mol Ecol 19:1774–1786

Zuur AF, Ieno EN, Walker NJ, Saveliev AA, Smith GM (2009) Mixed Effects Models and Extensions in Ecology with R. Springer, NY