

Date received: May 24, 2022; Date revised: August 16, 2022; Date accepted: August 23, 2022

DOI: <https://dx.doi.org/10.4314/sinet.v45i2.7>

Optimization of *Neochetina eichhorniae* Warner and *Neochetina bruchi* Hustache for the management of water hyacinth, *Eichhornia crassipes* (Mart.) Solms in the Central Rift Valley lakes through the use of different nutrient levels in Ethiopia

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ABSTRACT: Water hyacinth, *Eichhornia crassipes* (Mart.) Solms, is an alien invasive weed in Ethiopia that has been established in freshwater bodies. *Eichhornia crassipes* negatively affects water transportation, fishing and biodiversity among others. Hence, implementation of eco-friendly management option such as biological control is a mandatory. The weevils, *Neochetina eichhorniae* Warner and *N. bruchi* Hustache, are the principal biocontrol agents against *E. crassipes* in their native ranges and have been introduced to Ethiopia for the management of the weed. The weevils and their host are affected by the nitrate and phosphate contents of the water body. Thus, this study was conducted to investigate the effect of *N. bruchi* and *N. eichhorniae* in relation to water nutrient contents in the management of *E. crassipes*. The experiment was conducted in a randomized complete block design with three replications in a 4x4x3 factorial arrangement. Eight weeks after the treatments' application, the population of the weevils and *E. crassipes* growth parameters were recorded. The larval population was significantly ($p < 0.05$) affected only by the interaction effect of *N. bruchi* and the nutrient levels. Among the treatments, the highest mean number of larvae was found on the plants given high and medium nutrient levels. There was also a significant interaction ($p < 0.05$) between the two weevils adult density. The mean number of emerged adult density of the weevils increased with increasing nutrient levels. The nutrients and weevils exhibited a significant interaction effect on *E. crassipes* leaf scarring and petiole tunnel. Leaf scarring at the highest weevil density and nutrient level was sixfold greater than that at the lower nutrient level and weevil density. The tunnel length of the weed was threefold higher at the highest nutrient level regardless of the weevil density. The growth parameters of *E. crassipes* were found to be greater at high nutrient level, except for the mean number of flowers. Conversely, the growth parameters showed a non-uniform trend with the increasing density of the weevils. The current result suggests reduction in *E. crassipes* could be achieved by lowering the water nutrient levels and use of *N. bruchi* and *N. eichhorniae*.

Key words/phrases: Biological control, Freshwater, Weeds, Weevils

INTRODUCTION

Water hyacinth (*Eichhornia crassipes* (Mart.) Solms (Pontederiaceae)) is a free-floating macrophyte that invades fresh water and reproduces sexually as well as vegetatively. *Eichhornia crassipes* is native to South America, with the suggestion that the center of origin may be the upper reaches of the Amazon River and its tributaries (Gopal, 1987; Julien, 2000). *Eichhornia crassipes* is known to have been introduced into tropical and temperate regions as an ornamental plant due to its beautiful flower (Little, 1965; Cilliers *et al.*, 2003). Currently, it is among the aquatic weeds, especially problematic in Africa. In Ethiopia, the weed is causing a

devastating problem by affecting biodiversity, water transportation and fishing among others in the water bodies of the Rift Valley and Lake Tana (Firehun Yirefu *et al.*, 2014; Adugnaw Admas *et al.*, 2017). A recent report showed that *E. crassipes* covered approximately 2% of Lake Tana, reducing its potential for fish production by 92% (Nagassa Dechassa and Belay Abate, 2020). Its infestation in the Central Rift Valley was reported in the 1950s, but attained pest status very recently, mainly because of intensified agricultural systems around water bodies that discharged nutrients into the system. A good example is Lake Zuwayi which currently invaded by *E. crassipes* (Emana Getu, unpublished data).

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For the management of *E. crassipes*, physical and chemical control have been used extensively around the world to get short-term control (Dereje Tewabe, 2015; LVEMP, 2016), while wetland management and biological control were used as a long-term control (Harley *et al.*, 1996; Julien, 1997; Mumma, 1999). Rapid proliferation of *E. crassipes* was observed in eutrophic water bodies (Yu *et al.*, 2019) where its natural enemies were absent (O'Brien, 1976; Njoka, 2004; LVEMP, 2006). *Neochetina eichhorniae* Warner and *N. bruchi* Hustache (Order: Coleoptera, Family: Curculionidae) are among the main biocontrol agents found to be popular and effective in the management of *E. crassipes* (Mumma, 1999; Julein, 2001).

The biocontrol agents are native to the Amazon basin in South America, along with *E. crassipes* (Manning, 1979). In 1972, *N. eichhorniae* was the first insect approved for release as a biocontrol agent of *E. crassipes* followed by *N. bruchi* in 1974 on canals and lakes of Fort Lauderdale, Florida (O'Brien, 1976). These species are now distributed in over 30 countries in the world throughout the distribution range of *E. crassipes* as a classical biological control (Firehun Yirefu *et al.*, 2016). This approach was kicked off in Ethiopia following the rapid expansion and proliferation of *E. crassipes* in newly infested water bodies (Wassie Anteneh *et al.*, 2015; Firehun Yirefu *et al.*, 2016; Adugnaw Admas *et al.*, 2017).

Biotic potential of *N. eichhorniae* and *N. bruchi* is influenced by a variety of environmental factors such as temperature, water nutrients and herbicide use among others (Wilson *et al.*, 2006; Wise *et al.*, 2007). *Neochetina eichhorniae* and *N. bruchi* were imported from Uganda and kept under quarantine at Ethiopian Sugar Corporation, Research and Development Center, Wonji (Firehun Yirefu *et al.*, 2015). Under controlled condition, adaptability test was conducted and the biocontrol agents found to be suitable for the area. There were no variables studied in relation to their potential such as the effect of water nutrient levels both on the biocontrol agents and *E. crassipes*. Experiences elsewhere indicated that the efficacy of the biocontrol agents vary from place to place that implies the need of optimization to exploit the

maximum benefit from the release of the weevils in the water body. Hence, the current study was conducted to understand the effect of water nutrient levels, *N. eichhorniae* and *N. bruchi* densities on *E. crassipes*.

MATERIALS AND METHODS

Description of the study area

The study was conducted from April 2020 to October 2020 in the Lath House at Ethiopian Sugar Corporation, Research and Development Center, Wonji. Wonji is located approximately 67 km southeast of Addis Ababa. It is positioned in the central part of the main East African Rift Valley at 8°30' - 8°35' N and 39°20' E and at an altitude of 1540 meters above sea level. The annual precipitation, mean maximum, and minimum temperature of the area were 831 mm, 27°C, and 15°C, respectively (Firehun Yirefu *et al.*, 2015). The area has benefited from Awash River irrigation-based agricultural practices. The Awash River is one of the major rivers of Ethiopia that is highly utilized and the first basin to be introduced to modern agriculture (Fekadu Aduna *et al.*, 2021). It starts from the Highlands of Ginchi, flows toward the east and north of the country, and ends in the country after a long journey of approximately 1,200 km (Addis Anteneh and Hailu Yemanu, 1970).

Cultivation of Eichhornia crassipes

Uniform and healthy *E. crassipes* plants with no symptoms of disease were collected for the experiment (Dissanayake *et al.*, 2009). *Eichhornia crassipes* collection was done from Camp 9 reservoir, located approximately 10 km southeast of the Wonji town. Four healthy young *E. crassipes* plants with three to four newly emerged leaves were placed in a plastic bucket of 15 liter capacity with a size of 16 cm in height x 40 cm in width and acclimatized for a week prior to the start of the experiment following Gore (2017). Each plastic bucket containing *E. crassipes* was treated with nitrate (KNO₃) and phosphate (KH₂PO₄) sources (Bownes, 2008) at different levels. The levels used were low (10 mg/l N, 0.18 mg/l P), medium (35 mg/l N, 1.68 mg/l P), and high (100 mg/l N, 3 mg/l P) following a survey of water nutrient levels detected in fresh water bodies in Ethiopia by

Andualem Mekonnen *et al.* (2014) and Yirga Kebede (2016). Once the experiment commenced, the fertilizers were applied every week after replacing the water throughout the study period following Reddy *et al.* (1989).

Rearing of *Neochetina bruchi* and *Neochetina eichhorniae*

For both *N. bruchi* and *N. eichhorniae*, 150 adults each were collected from the quarantine lath house and mass reared separately in a new lath house at Ethiopian Sugar Corporation Research and Development Center, Wonji. Rearing was done on *E. crassipes* kept in round plastic buckets of 20 liter capacity with a size of 25 cm in height x 55 cm in width filled with water to $\frac{3}{4}$ and contained in rearing cages (Firehun Yirefu *et al.*, 2016). The rearing cages were made of insect proof polyethylene mesh with a size of 50 cm in width and 90 cm in height, exposed to a 12:12 photoperiod. The temperature and relative humidity of the lath house in which the cages were placed were 17.1°C to 33.7°C and 22% to 83%, respectively. Newly emerged five day-old *N. bruchi* and *N. eichhorniae* were collected separately. The adult weevils were grouped into male and female individuals per species to maintain their sex ratio for experiment by using keys of dimorphism such as snout length, snout curviness, and shininess of the snout tip (Dissanayake *et al.*, 2009; Ray, 2015).

Treatments and experimental design

In this experiment, 144 round plastic buckets with a size of 16 cm in height x 40 cm in width were used, in which each bucket was considered as a plot. The experimental design consisted of three rows of plastic buckets, each with 0.3 m between plots and 0.1 m between rows. The experiment was designed in a randomized complete block design (RCBD) in a 4x4x3 factorial arrangement in three replications. *Neochetina bruchi* and *N. eichhorniae* each with four density levels were combined with three nutrient levels to make factorial treatments (Center *et al.*, 1982). The density levels of the two weevils were zero, one pair, two pairs, and three pairs following Firehun Yirefu *et al.* (2016), while the *E. crassipes* plant nutrient levels were low (10 mg/1 N, 0.18 mg/1 P), medium (35 mg/1 N, 1.68 mg/1 P) and high (100 mg/1 N, 3 mg/1 P) as described in the *E. crassipes* cultivation methodology. *Eichhornia crassipes* plants contained

in round plastic buckets were subjected to different nutrient concentration levels for a week and subsequently treated with the weevils (Mailu, 2001). Once the treatment application was commenced, each plastic bucket containing *E. crassipes* that received the treatments was set in insect proof cages to prevent the entrance of other herbivores and natural enemies of the weevils following DeLoach (1976).

Data collection

Eight weeks after treatment application, data on live leaf number, leaf loss, ramet number, and flower number of *E. crassipes* per plot were collected (Heard and Winterton, 2000; Mukarugwiro *et al.*, 2018). Leaf scars, length, and diameter per lamina were counted from randomly selected 3rd lamina of a *E. crassipes* inside out, while petiole tunnel, length and diameter per plant were measured from the longest petiole per a *E. crassipes* plant (Jones *et al.*, 2018).

Eichhornia crassipes biomass and the density of *N. bruchi* and *N. eichhorniae* were recorded three months after treatment application (Goyer and Stark, 1984). *Eichhornia crassipes* fresh weight was taken soon after the sample was blotted; while dry weight was taken after the samples were oven dried for 27°C hrs at 75°C following Bock (1969).

Two petioles with tunnels were randomly selected from each of the three *E. crassipes* plants per plot for recording larvae of both *N. bruchi* and *N. eichhorniae*, and visible larval stages (2nd and 3rd larvae) were counted after petiole dissection (Jadhav *et al.*, 2008). Adult *N. bruchi* and *N. eichhorniae* were counted from four randomly selected *E. crassipes* plants per plot by examining the individual plants (Pratiwi *et al.*, 2018).

Statistical analysis

Data collected on *E. crassipes*, *N. eichhorniae*, and *N. bruchi* parameters were subjected to analysis of variance using the SAS 9.4 software package. Transformations were carried out for data that were not normally distributed. The logarithmic transformation was used for leaf length, leaf diameter, and tunnel length. Data on larva, adult, leaf scar, dry weight, petiole diameter, and leaf loss were normalized by the inverse distribution function, while leaf number data were transformed using square root transformation. The strength of the linear relationship within the *E. crassipes*

growth parameters was determined by two tailed Pearson correlation. Mean and standard error were used from descriptive statistics, and significant means were separated using Tukey's studentized range (HSD) at 5%. The significance and non-significance values of the tests in the results were denoted by $p < 0.05$ and $p \geq 0.05$, respectively.

RESULTS AND DISCUSSION

Effect of nutrient levels, Neochetina bruchi and Neochetina eichhorniae densities on larval emergence

The interaction effect was only significant ($p < 0.05$) with *N. bruchi* and nutrient levels for larval density. *Eichhornia crassipes* plants that received medium and higher nutrient levels exhibited higher larval density of the weevils compared to the plants which received low nutrient level (Table 1). The current result is in line with Heard and Winterton (2000), who reported that *N. bruchi* and *N. eichhorniae* fecundity and immature stage survival were higher at higher nutrient level (1.6 mg /l N and 1.0 mg/l P). The highest mean

number of larvae (15.3) was recorded on *E. crassipes* plants that received the treatment combinations of the highest density of weevils with high nutrient level. This result was significantly ($p < 0.05$) different from the lowest mean number (4.0) obtained with the treatment combination of *N. eichhorniae* and the lowest water nutrient (Table 1). In contrast to the current findings, Wilson *et al.* (2006) reported that the larval stage mortality of weevils increased and that their density became low on *E. crassipes* plants, where the initial density of weevils was high regardless of nutrient levels.

In the main effect of adult *N. eichhorniae*, the highest larval mean number (9.11) was recorded on *E. crassipes* plants that received the highest density of weevils. *Eichhornia crassipes* plants subjected to treatments that incorporated *N. eichhorniae* adults resulted in a higher larval mean number compared to the untreated control (Figure 1). This result is in agreement with Julien *et al.* (1999) who reported that *N. eichhorniae* density was not sensitive to *E. crassipes* quality, unlike *N. bruchi* density (Center and Dray, 1992; Ray, 2015).

Table 1. Effect of nutrient levels, *N. bruchi* and *N. eichhorniae* densities on larva emergence

Treatment	Mean±SE	Treatment	Mean±SE	Treatment	Mean±SE
b0 e1 p1	5.3±0.7 ^{bdc}	b1 e2 p1	6.7±1.7 ^{bdc}	b2 e3 p1	5.3±1.3 ^{bdc}
b0 e1 p2	6.3±0.3 ^{bdac}	b1 e2 p2	7.7±1.8 ^{bdac}	b2 e3 p2	10.3±2.4 ^{bdac}
b0 e1 p3	7.7±1.2 ^{bdac}	b1 e2 p3	8.0±1.0 ^{bdac}	b2 e3 p3	12.7±1.7 ^{bac}
b0 e2 p1	4.0±1.9 ^d	b1 e3 p1	8.0±2.0 ^{bdac}	b3 e0 p1	5.0±2.0 ^{bdc}
b0 e2 p2	5.3±0.9 ^{bdc}	b1 e3 p2	8.7±1.2 ^{bdac}	b3 e0 p2	10.3±0.9 ^{bdac}
b0 e2 p3	5.7±1.2 ^{bdc}	b1 e3 p3	10.0±1.5 ^{bdac}	b3 e0 p3	10.3±1.3 ^{bdac}
b0 e3 p1	5.0±0.6 ^{bdc}	b2 e0 p1	6.7±1.7 ^{bdc}	b3 e1 p1	5.0±1.5 ^{bdc}
b0 e3 p2	5.7±0.7 ^{bdc}	b2 e0 p2	8.3±1.8 ^{bdac}	b3 e1 p2	8.7±2.2 ^{bdac}
b0 e3 p3	6.3±0.9 ^{bdac}	b2 e0 p3	9.0±1.7 ^{bdac}	b3 e1 p3	11.0±2.0 ^{bdac}
b1 e0 p1	6.0±2.0 ^{bdc}	b2 e1 p1	6.0±1.0 ^{bdc}	b3 e2 p1	5.3±0.7 ^{bdc}
b1 e0 p2	7.0±1.5 ^{bdac}	b2 e1 p2	8.0±1.5 ^{bdac}	b3 e2 p2	13.0±2.1 ^{bac}
b1 e0 p3	8.7±1.2 ^{bdac}	b2 e1 p3	8.7±1.9 ^{bdac}	b3 e2 p3	13.3±1.2 ^{ba}
b1 e1 p1	6.0±1.5 ^{bdc}	b2 e2 p1	4.3±1.5 ^{dc}	b3 e3 p1	7.0±1.7 ^{bdac}
b1 e1 p2	8.7±2.2 ^{bdac}	b2 e2 p2	10.7±1.7 ^{bdac}	b3 e3 p2	15.0±1.5 ^a
b1 e1 p3	8.7±1.2 ^{bdac}	b2 e2 p3	11.3±1.3 ^{bdac}	b3 e3 p3	15.3±0.9 ^a

Letters under column "Treatment" refer to weevils (b = *N. bruchi*, e = *N. eichhorniae*) and plants (p = *E. crassipes* plant), while numbers associated with the letters refer to densities (0 = control, 1 = one pair, 2 = two pairs, and 3 = three pairs) and nutrient concentrations (1 = low, 2 = medium, and 3 = high) of the weevils and *E. crassipes*, respectively. In columns, SE = standard error. Mean values followed by the same letter(s) indicate no significant difference at the 5% level, Tukey's studentized range test (HSD).

Effect of nutrient levels, Neochetina bruchi and Neochetina eichhorniae densities on adult population

A significant interaction was found between *N. bruchi* and *N. eichhorniae* on their adult density ($p < 0.05$). *Eichhornia crassipes* plants in experimental

buckets had a high adult density of *N. bruchi* and *N. eichhorniae* when they were treated with a high density of the weevils. The highest mean number (43) was obtained using a high adult density of weevils on *E. crassipes* plants that received the highest nutrient level (Table 2), which was

substantially different from the lowest result. In a combined application of the weevil with white amur, Delfosse *et al.* (1976) found a maximum of 89.4 *N. eichhorniae* per 388 *E. crassipes* plants in a pool per season, which is lower than the current result.

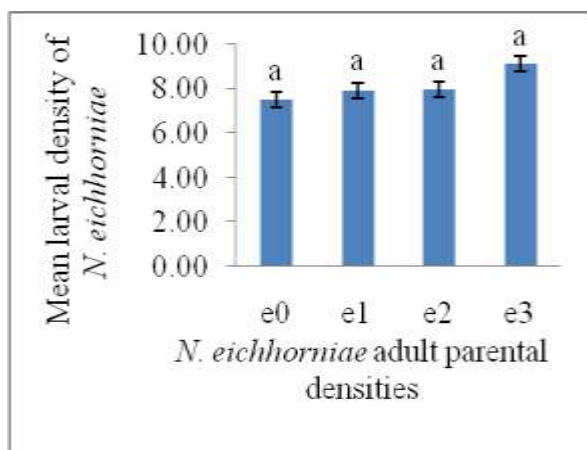


Figure 1. Mean larval density of *N. eichhorniae* (progeny) at different adult parental density of *N. eichhorniae*, e0-e3 (e= *N. eichhorniae*, e0= control, e1= one pair, e2= two pairs, and e3= three pairs). Means sharing the same letter are not significantly different from each other at the 5% level, Tukey’s studentized range test (HSD).

In the main effect, the mean density of adult weevils recorded from *E. crassipes* plants treated with a high nutrient level was the highest, and it

was significantly different from the medium and untreated control (Figure 2). This is consistent with the work of Center and Dray (2010), who found that a high nutrient level resulted in a higher density of weevils in *E. crassipes* plants when treated with a higher combination of *N. eichhorniae* and *N. bruchi* density than when treated alone. Similarly, Ismail *et al.* (2017) reported that *E. crassipes* subjected to high nutrient level had the greatest adult mean population compared to other treatments.

In comparison to those cultivated at an oligotrophic level, *E. crassipes* plants grown at a greater nutrient levels were preferred hosts for *N. bruchi*, but not for *N. eichhorniae* (Kidd, 2000). In this situation, *N. bruchi* generated a greater number of progeny, which severely destroyed the *E. crassipes* (Heard and Winterton, 2000). According to a report by Bick *et al.* (2020), salinity and nutrients that influenced *E. crassipes* had a significant impact on *N. bruchi*'s life cycle. A factor that has a substantial negative impact on the biocontrol agents has a disproportional detrimental impact on the host plant (Wilson *et al.*, 2005). As a result, a number of ecological processes must be integrated to reduce the weed status in a given area and eventually eradicate it (Hopper *et al.*, 2021).

Table 2. Effect of nutrient levels, *N. bruchi* and *N. eichhorniae* densities on adult

Treatment	Mean±SE	Treatment	Mean±SE	Treatment	Mean±SE
b0 e1 p1	13.7±2.3 ehgf	b1 e2 p1	15.7±1.9 edhgef	b2 e3 p1	25.3±1.5 ebdacf
b0 e1 p2	17.0±2.5 edhgef	b1 e2 p2	20.0±3.8 ebdhgef	b2 e3 p2	32.7±5.8 bdac
b0 e1 p3	17.3±2.2 ebdhgef	b1 e2 p3	22.7±2.9 ebdhgef	b2 e3 p3	41.3±1.3 a
b0 e2 p1	15.0±2.5 edhgf	b1 e3 p1	15.7±1.5 edhgef	b3 e0 p1	11.0±3.5 hg
b0 e2 p2	16.3±2.2 edhgef	b1 e3 p2	24.0±0.6 ebdagef	b3 e0 p2	11.0±3.2 hg
b0 e2 p3	17.0±2.1 edhgef	b1 e3 p3	29.3±1.2 ebdac	b3 e0 p3	16.3±4.2 edhgef
b0 e3 p1	14.3±1.9 edhgf	b2 e0 p1	15.0±2.1 edhgf	b3 e1 p1	10.3±2.3 h
b0 e3 p2	15.7±2.0 edhgef	b2 e0 p2	18.7±0.3 ebdhgef	b3 e1 p2	16.3±2.0 edhgef
b0 e3 p3	17.7±1.9 ebdhgef	b2 e0 p3	18.7±0.3 ebdhgef	b3 e1 p3	19.7±3.8 ebdhgef
b1 e0 p1	15.3±1.8 edhgef	b2 e1 p1	18.0±2.1 ebdhgef	b3 e2 p1	22.7±3.8 ebdhgef
b1 e0 p2	16.7±0.7 edhgef	b2 e1 p2	26.7±3.5 ebdacf	b3 e2 p2	39.7±4.8 a
b1 e0 p3	20.0±4.4 ebdhgef	b2 e1 p3	33.0±2.1 bdac	b3 e2 p3	40.0±1.7 ba
b1 e1 p1	15.0±2.1 edhgf	b2 e2 p1	27.0±3.2 ebdacf	b3 e3 p1	34.0±3.8 bac
b1 e1 p2	19.3±2.0 ebdhgef	b2 e2 p2	34.3±1.9 bac	b3 e3 p2	41.7±1.5 a
b1 e1 p3	25.0±1.7 ebdacf	b2 e2 p3	40.3±2.3 a	b3 e3 p3	43.0±2.5 a

Letters under column "Treatment" refer to weevils (b= *N. bruchi*, e= *N. eichhorniae*) and plants (p= *E. crassipes* plant), while numbers associated with the letters refer to the densities (0= control, 1= one pair, 2= two pairs and 3= three pairs) and nutrient concentrations (1= low, 2= medium and 3= high) of the weevils and *E. crassipes*, respectively. On columns, SE=standard error. Mean values followed by the same letter(s) indicate no significant difference at the 5% level, Tukey’s studentized range test (HSD).

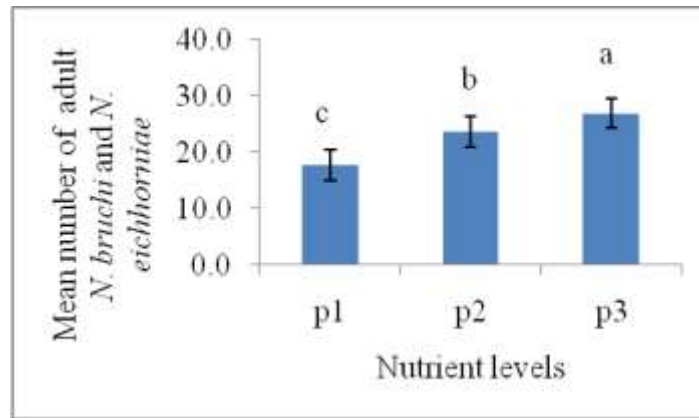


Figure 2. Adult *N. bruchi* and *N. eichhorniae* at different nutrient levels, p1-p3 (p= *E. crassipes* plant, p1= low, p2= medium, and p3=high). Means sharing the same letter are not significantly different from each other at the 5% level, Tukey's studentized range test (HSD).

Eichhornia crassipes leaf scar damage by *Neochetina bruchi* and *Neochetina eichhorniae*

Neochetina bruchi, *N. eichhorniae*, and *E. crassipes* nutrient levels had a significant interaction on leaf scarring ($p < 0.05$). *Eichhornia crassipes* plants that received the treatment combination of low *N. eichhorniae* density and poor nutrient with no *N. bruchi* had the lowest mean (9.0 per lamina) *E. crassipes* leaf scar (Table 3). The treatment combination with the highest weevil density and high nutrient had the highest mean (53.7 per lamina), which was significantly different from all other treatments (Table 3). The highest feeding scar in this study (53.7 per lamina) was lower than the previous study by Wright and Center (1984) who

recorded 91 scars per lamina, but higher than 11.1 and 24.66 scars per lamina by a single insect in 24 hrs for *N. bruchi* and *N. eichhorniae*, respectively (Gore *et al.*, 2020). Scars were enhanced by increasing the density of weevils and the nutrient content of *E. crassipes* (Hopper *et al.*, 2021). In another study (Heard and Winterton, 2000), adult feeding scars were unaffected by nutrient concentrations, but the presence of old leaf scars on *E. crassipes* hindered adult weevil feeding performance (Buchanan, 2013). However, when both weevils were combined, the injury to *E. crassipes* increased (DeLoach and Cordo, 1976; Gupta and Yadav, 2020).

Table 3. *Neochetinia bruchi* and *N. eichhorniae* feeding scars per leaf of *E. crassipes*.

Treatment	Mean+SE	Treatment	Mean+SE	Treatment	Mean+SE
b0 e1 p1	9.0±1.5 ^r	b1 e2 p1	33.2±2.6 ^{ihgfi}	b2 e3 p1	26.1±1.8 ^{nml}
b0 e1 p2	16.7±1.5 ^{pq}	b1 e2 p2	35.1±3.5 ^{iehgfi}	b2 e3 p2	28.7±1.8 ^{knmlj}
b0 e1 p3	17.9±1.4 ^{opq}	b1 e2 p3	35.3±2.0 ^{ehgf}	b2 e3 p3	33.3±2.4 ^{ihgfi}
b0 e2 p1	15.3±1.5 ^q	b1 e3 p1	30.8±1.9 ^{ikhij}	b3 e0 p1	22.5±1.7 ^{onp}
b0 e2 p2	15.5±1.5 ^q	b1 e3 p2	39.2±3.1 ^{edf}	b3 e0 p2	23.3±1.9 ^{onm}
b0 e2 p3	35.8±1.5 ^{ehgf}	b1 e3 p3	39.8±3.4 ^{ed}	b3 e0 p3	33.5±1.7 ^{iehgfi}
b0 e3 p1	17.1±1.4 ^{pq}	b2 e0 p1	31.7±1.5 ^{ikhgij}	b3 e1 p1	18.1±2.3 ^{opq}
b0 e3 p2	27.1±1.4 ^{knml}	b2 e0 p2	31.8±2.5 ^{ikhgij}	b3 e1 p2	25.7±2.7 ^{nml}
b0 e3 p3	31.7±1.5 ^{ikhgij}	b2 e0 p3	43.7±2.4 ^{cd}	b3 e1 p3	34.1±2.3 ^{iehgfi}
b1 e0 p1	15.8±2.1 ^q	b2 e1 p1	22.1±1.2 ^{onp}	b3 e2 p1	26.0±2.1 ^{knml}
b1 e0 p2	29.5±2.4 ^{ikmlj}	b2 e1 p2	23.9±2.3 ^{onm}	b3 e2 p2	38.3±3.8 ^{egf}
b1 e0 p3	29.8±2.1 ^{ikmlj}	b2 e1 p3	32.0±1.7 ^{ikhgj}	b3 e2 p3	51.7±1.7 ^b
b1 e1 p1	11.1±1.2 ^r	b2 e2 p1	32.9±1.8 ^{ihgfi}	b3 e3 p1	29.3±3.8 ^{ikmlj}
b1 e1 p2	15.8±2.1 ^q	b2 e2 p2	34.9±3.0 ^{iehgfi}	b3 e3 p2	34.6±3.8 ^{iehgfi}
b1 e1 p3	18.9±2.8 ^{opq}	b2 e2 p3	50.0±2.3 ^{cb}	b3 e3 p3	53.7±1.3 ^a

Letters under column "Treatment" refer to weevils (b = *N. bruchi*, e = *N. eichhorniae*) and plants (p = *E. crassipes* plant), while the numbers associated with the letters refer to the densities (0=control, 1= one pair, 2= two pairs and 3= three pairs) and nutrient concentrations (1= low, 2= medium and 3= high) of the weevils and *E. crassipes*, respectively. On columns, SE=standard error. Mean values followed by the same letter(s) indicate no significant difference at the 5% level, Tukey's studentized range test (HSD).

Eichhornia crassipes petiole tunnel damage by *Neochetina bruchi* and *Neochetina eichhorniae*

The interaction of *E. crassipes* weevils had a significant effect on the tunnel of the *E. crassipes* petiole ($p < 0.05$). The treatment combination of no *N. bruchi*, low *N. eichhorniae* density, and low nutrient level resulted in the lowest mean (1.2 cm per plant) of *E. crassipes* tunnel length, which was significantly different from all other treatments (Table 4). On *E. crassipes* with high nutrient level but varied density of weevils, the highest mean of tunnel (4.5 cm per plant) was recorded (Table 4). This demonstrated that tunnel length maximized since damage was proportional to the amount of *E. crassipes* nutrient present, regardless of density.

Wilson *et al.* (2005) found that at the highest nutrient level, *N. eichhorniae* larvae grew larger and matured faster, resulting in more damage. This was in contrast to Mukarugwiro *et al.* (2018), who found that *E. crassipes* plants kept at low nutrient level had the highest *N. eichhorniae* larval feeding rates. This meant that increasing *E. crassipes* nutrient levels had a detrimental impact on the weevils' ability to damage the plant (Moran, 2006). The damage intensity increased as the density of weevils increased (Hopper *et al.*, 2017). The current study's findings were supported by Firehun Yirefu *et al.* (2016), who found that the weevils' interaction effect was better for the *E. crassipes* tunnel than when they were alone.

Table 4. *Neochetinia bruchi* and *N. eichhorniae* tunnel (cm) on the petiole of *E. crassipes*.

Treatment	mean ± SE	Treatment	mean ± SE	Treatment	mean ± SE
b0 e1 p1	1.2±0.15 ^m	b1 e2 p1	2.4±0.09 ^{hfig}	b2 e3 p1	2.5±0.12 ^{hefig}
b0 e1 p2	3.8±0.18 ^{bac}	b1 e2 p2	3.2±0.09 ^{edc}	b2 e3 p2	2.7±0.18 ^{hefdg}
b0 e1 p3	4.4±0.17 ^a	b1 e2 p3	4.4±0.12 ^a	b2 e3 p3	3.4±0.15 ^{bdc}
b0 e2 p1	1.6±0.23 ^l	b1 e3 p1	2.4±0.12 ^{hfigk}	b3 e0 p1	1.9±0.09 ^{jlk}
b0 e2 p2	2.2±0.20 ^{hijk}	b1 e3 p2	2.5±0.09 ^{hefig}	b3 e0 p2	3.5±0.09 ^{bdc}
b0 e2 p3	2.5±0.06 ^{hefig}	b1 e3 p3	3.0±0.09 ^{efdc}	b3 e0 p3	4.1±0.18 ^a
b0 e3 p1	2.2±0.12 ^{hijk}	b2 e0 p1	1.7±0.15 ^l	b3 e1 p1	2.3±0.09 ^{hijk}
b0 e3 p2	2.4±0.13 ^{hijk}	b2 e0 p2	2.5±0.15 ^{hefig}	b3 e1 p2	3.2±0.18 ^{edc}
b0 e3 p3	3.0±0.03 ^{efdc}	b2 e0 p3	4.4±0.21 ^a	b3 e1 p3	3.4±0.06 ^{bdc}
b1 e0 p1	2.4±0.19 ^{hijk}	b2 e1 p1	1.9±0.06 ^{lk}	b3 e2 p1	2.9±0.07 ^{efdg}
b1 e0 p2	3.0±0.03 ^{efdc}	b2 e1 p2	3.3±0.15 ^{bdc}	b3 e2 p2	3.4±0.06 ^{bdc}
b1 e0 p3	3.1±0.09 ^{edc}	b2 e1 p3	4.2±0.15 ^a	b3 e2 p3	4.5±0.17 ^a
b1 e1 p1	2.2±0.12 ^{hijk}	b2 e2 p1	2.0±0.09 ^{jilk}	b3 e3 p1	2.4±0.06 ^{hfigk}
b1 e1 p2	2.9±0.07 ^{efdg}	b2 e2 p2	3.3±0.15 ^{bdc}	b3 e3 p2	2.9±0.06 ^{efdg}
b1 e1 p3	3.9±0.18 ^{ba}	b2 e2 p3	4.3±0.15 ^a	b3 e3 p3	3.8±0.12 ^{bac}

Letters under column "Treatment" refer to weevils (b= *N. bruchi*, e= *N. eichhorniae*) and plants (p= *E. crassipes* plant), while numbers associated with the letters refer to the densities (0= control, 1= one pair, 2= two pairs, and 3= three pairs) and nutrient concentrations (1= low, 2= medium, and 3= high) of the weevils and *E. crassipes*, respectively. On columns, SE=standard error. Mean values followed by the same letter(s) indicate no significant difference at the 5% level, Tukey's studentized range test (HSD).

Effect of water nutrient levels, *Neochetina bruchi* and *Neochetina eichhorniae* densities on the biomass of *Eichhornia crassipes*

The interaction of nutrients with *N. bruchi* or *N. eichhorniae* had no significant effect on *E. crassipes* fresh weight. In the fresh weight measurements, the maximum mean was obtained at high water nutrient level, whereas the lowest mean was obtained from an untreated control (Figure 3A). The mean fresh weight of *E. crassipes* grown at the highest nutrient level differed significantly from that of the untreated control and medium nutrient levels, but untreated control and medium nutrient

levels showed no significant difference between each other (Figure 3A).

This indicates that ecological factors such as nutrients play a role in *E. crassipes* biomass (Voukeng, 2017). *Neochetina bruchi* and *N. eichhorniae*, on the other hand, had an effect on the fresh weight of *E. crassipes*. The fresh weight showed an increasing trend with a decreasing population density in both weevils (Figure 3B and C). A similar trend was observed in postrelease evaluation in the Chittar River, India, where weevil density increased after inoculation and slowed down *E. crassipes* biomass as a result of

stunted growth and low reproduction (Sivaraman and Murugesan, 2017).

The interaction of the two weevils and the *E. crassipes* nutrient levels resulted in a significant effect ($p < 0.05$) on the dry weight of *E. crassipes*. In the current study, the highest mean (2.70 g) *E. crassipes* dry weight was found in the untreated control with the treatment combination of high water nutrient level and free of weevils (Table 5).

On the other hand, the treatment combination of the highest weevils' density and the highest water nutrient level resulted in the lowest mean (2.46 g) *E. crassipes* dry weight (Table 5). This indicates that the dry weight of *E. crassipes* plants was influenced by weevils' density (Firehun Yirefu *et al.*, 2016) and water nutrient levels, which had a greater impact than their herbivores (Coetzee and Hill, 2012).

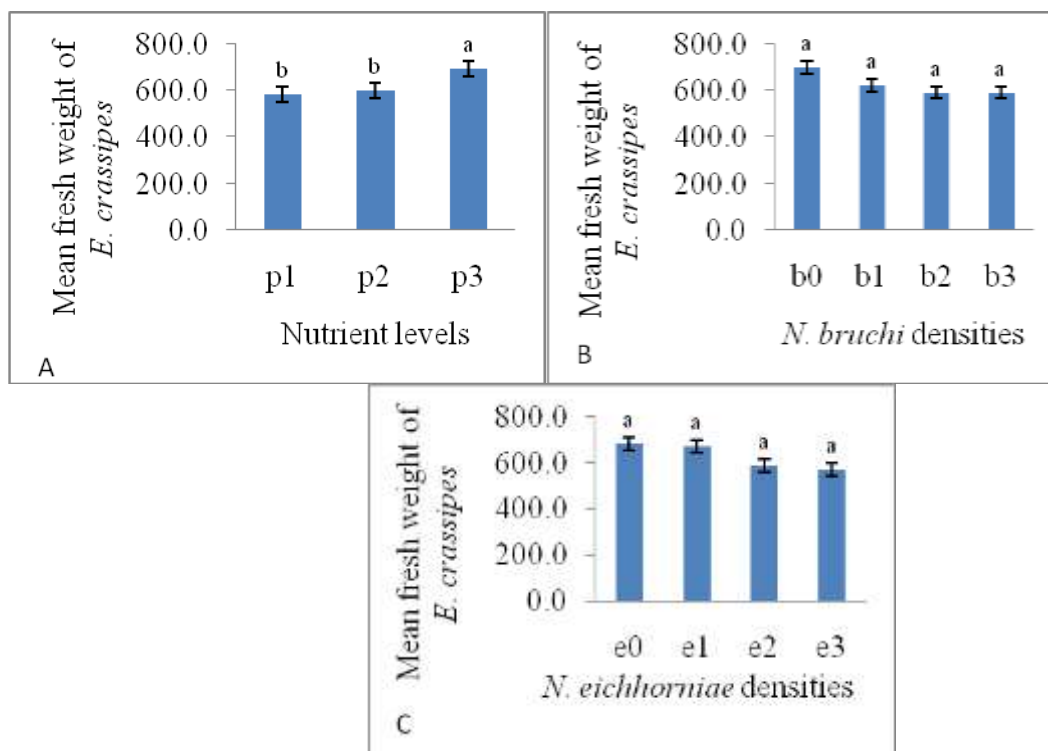


Figure 3. Effect of nutrient concentration levels, p1-p3 (p= *E. crassipes* plant on different nutrient concentration levels, p1= low, p2= medium, and p3= high) on *E. crassipes* fresh weight (A), effect of *N. bruchi* densities, b0-b3 (b= *N. bruchi*, b0= control, b1= one pair, b2= two pairs, and b3= three pairs) on *E. crassipes* fresh weight (B) and effect of *N. eichhorniae* densities, e0-e3 (e= *N. eichhorniae*, e0= control, e1= one pair, e2= two pairs, and e3= three pairs) on *E. crassipes* fresh weight (C). Means sharing the same letter are not significantly different from each other at the 5% level, Tukey's studentized range test (HSD).

Effect of water nutrient levels, Neochetina bruchi and Neochetina eichhorniae densities on the length and diameter of Eichhornia crassipes leaves and petioles

There was nonsignificant ($p > 0.05$) interaction among *N. bruchi*, *N. eichhorniae* and water nutrient levels on *E. crassipes* leaf length. Weevils and water nutrient levels had a significant impact on *E. crassipes* leaf length (Figure 4A, B and C). With increasing densities of both *N. bruchi* and *N. eichhorniae*, the leaf length decreased dramatically (Figure 4A and B). The leaf length of *E. crassipes* grew longer at the highest nutrient than at the

lowest (Figure 4C). A significant interaction between *N. eichhorniae* and the water nutrient levels ($p < 0.05$) was revealed in the leaf diameter of the *E. crassipes*. *Eichhornia crassipes* leaf diameter was found to be small when nutrient level was low (Table 6). *Eichhornia crassipes* with the lowest nutrient level (untreated control treatment) subjected to the lowest density of weevils showed, the lowest mean (3.4 cm per plant) leaf diameter. On the other hand, regardless of weevils' species or density, the largest mean (7.2 cm per plant) leaf diameter was obtained on *E. crassipes* with the highest nutrient level (Table 6).

Table 5. Effect of nutrient levels, *N. bruchi* and *N. eichhorniae* densities on *E. crassipes* dry weight (g).

Treatment	Mean± SE	Treatment	Mean± SE	Treatment	Mean± SE
b0 e0 p1	2.53±0.01 mjlihkkn	b1 e1 p2	2.61±0.01 ebdghcf	b2 e2 p3	2.53±0.01 mjlihkkn
b0 e0 p2	2.66±0.01 bac	b1 e1 p3	2.61±0.01 ebdghcf	b2 e3 p1	2.53±0.02 mjlihkkn
b0 e0 p3	2.70±0.01 a	b1 e2 p1	2.61±0.01 ebdighcf	b2 e3 p2	2.58±0.01 ejdighcf
b0 e1 p1	2.65±0.01 ebdacf	b1 e2 p2	2.51±0.01 mlon	b2 e3 p3	2.54±0.01 mjlihkkn
b0 e1 p2	2.67±0.02 bac	b1 e2 p3	2.52±0.01 mjlkkn	b3 e0 p1	2.58±0.01 ejdighcf
b0 e1 p3	2.68±0.01 bac	b1 e3 p1	2.50±0.01 on	b3 e0 p2	2.59±0.01 ejdighcf
b0 e2 p1	2.53±0.02 mjlihkkn	b1 e3 p2	2.57±0.01 ejdighkf	b3 e0 p3	2.53±0.01 mjlihkkn
b0 e2 p2	2.57±0.01 ejdighkf	b1 e3 p3	2.57±0.02 eslighkf	b3 e1 p1	2.54±0.01 mjlihkkn
b0 e2 p3	2.52±0.02 mjlkkn	b2 e0 p1	2.58±0.01 ejdighcf	b3 e1 p2	2.64±0.01 ebdgcf
b0 e3 p1	2.53±0.01 mjlihkkn	b2 e0 p2	2.50±0.02 on	b3 e1 p3	2.69±0.01 ba
b0 e3 p2	2.57±0.02 ejdighkf	b2 e0 p3	2.63±0.01 ebdgcf	b3 e2 p1	2.63±0.01 ebdgcf
b0 e3 p3	2.60±0.01 ejdighcf	b2 e1 p1	2.57±0.01 ejdighkf	b3 e2 p2	2.59±0.02 ejdighcf
b1 e0 p1	2.53±0.01 mjlihkkn	b2 e1 p2	2.56±0.01 jlihkf	b3 e2 p3	2.51±0.01 mlon
b1 e0 p2	2.66±0.02 bac	b2 e1 p3	2.63±0.01 ebdgcf	b3 e3 p1	2.56±0.02 mjlihkkn
b1 e0 p3	2.64±0.01 ebdgcf	b2 e2 p1	2.53±0.01 mjlihkkn	b3 e3 p2	2.66±0.02 bac
b1 e1 p1	2.55±0.01 mjlihkkn	b2 e2 p2	2.55±0.01 mjlihkkn	b3 e3 p3	2.46±0.02 ^o

Letters under column "Treatment" refer to weevils (b= *N. bruchi*, e= *N. eichhorniae*) and plants (p= *E. crassipes* plant), while numbers associated with the letters refer to densities (0= control, 1= one pair, 2= two pairs and 3= three pairs) and nutrient concentrations (1= low, 2= medium and 3= high) of the weevils and *E. crassipes*, respectively. On columns, SE=standard error. Mean values followed by the same letter(s) indicate no significant difference at the 5% level, Tukey's studentized range test (HSD).

The *E. crassipes* leaf diameter was found to be low when *N. bruchi* density was high in the single effect of *N. bruchi*. The largest mean (5.44 cm) *E. crassipes* leaf diameter was recorded for the untreated control, whereas the smallest mean (4.78 cm) was obtained for the *E. crassipes* that received three pairs of *N. bruchi* (Figure 5). The mean leaf diameter of *E. crassipes* treated with two pairs and three pairs of *N. bruchi* was significantly ($p < 0.05$) different from the mean leaf diameter treated with one pair of *N. bruchi* and untreated weed (control). In this study, the reduction in mean leaf length and diameter was less than 1.5-fold (Figures 4, 5 and Table 6). This finding was lower than the 1.7- and 1.9-fold reductions in leaf length and diameter, respectively (Sivaraman and Murugesan, 2017). The leaf length and width reduction show slight differences since they may depend on the number of leaves present per *E. crassipes* plant.

The two weevils and the nutrient levels had a significant interaction on the petiole length of the *E. crassipes* ($p < 0.05$). *Eichhornia crassipes* petiole length and diameter were generally greater on the *E. crassipes* plant across high water nutrient level compared to the low nutrient level regardless of the weevils' densities (Tables 7 and 8). In this result, the *E. crassipes* petiole length and diameter reduction were twofold. Similar to the current study result, Coetzee *et al.* (2007) reported that the difference in petiole parameters of *E. crassipes* was doubled at high nutrient level relative to low nutrient level. The current study result was greater than the 50% reduction in the petiole parameters reported by Ajuonu *et al.* (2003) but lower than the 2.5- and 2.6-fold reductions reported by Sivaraman and Murugesan (2017) and Firehun Yirefu *et al.* (2016), respectively. These differences may be attributed to environmental or *E. crassipes* forms (long and slender to swollen or bulbous).

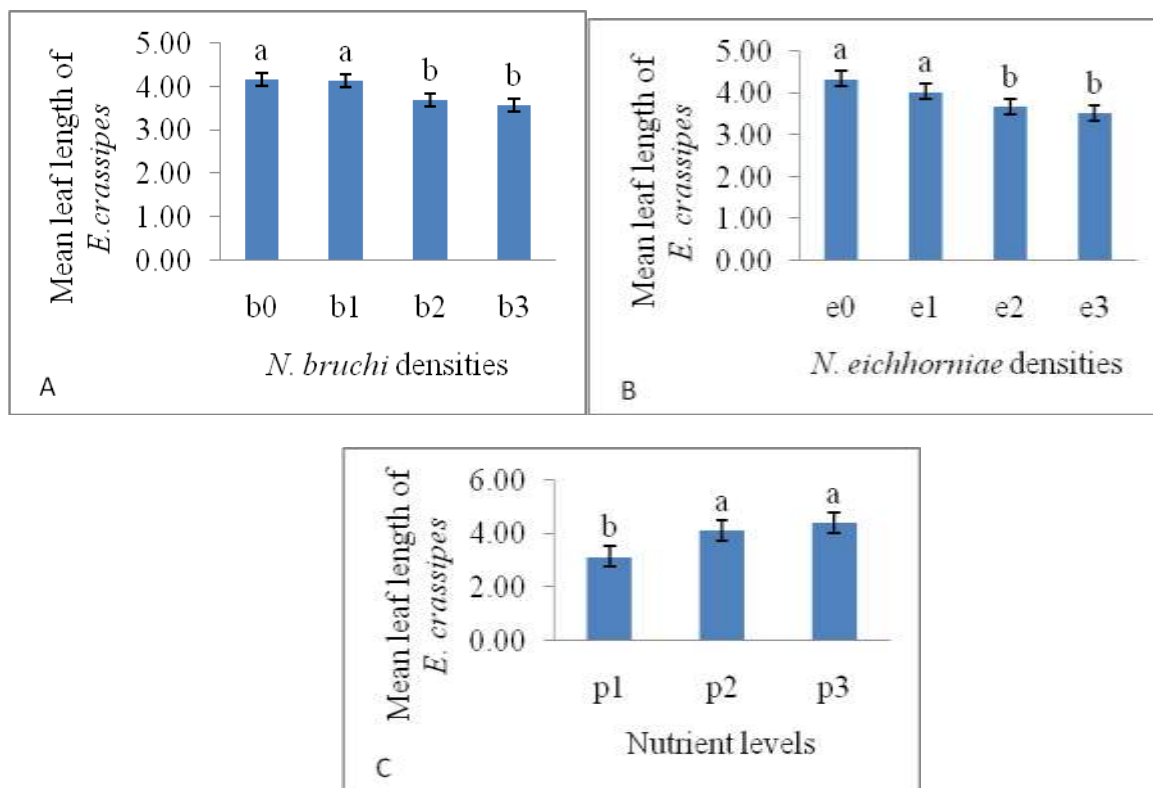


Figure 4. Effect of *N. bruchi* densities, b0-b3 (b= *N. bruchi*, b0= control, b1=one pair, b2= two pairs, and b3= three pairs) on *E. crassipes* leaf length (A), effect of *N. eichhorniae* densities, e0-e3 (e=*N. eichhorniae*, e0= control, e1= one pair, e2= two pairs, and e3= three pairs) on *E. crassipes* leaf length (B) and the effect of nutrient levels, p1-p3 (p= *E. crassipes* plant, p1= low, p2 = medium, and p3= high) on *E. crassipes* leaf length (C). Means sharing the same letter are not significantly different from each other at the 5% level, Tukey's studentized range test (HSD).

Table 6. Effect of nutrient levels, *N. bruchi* and *N. eichhorniae* densities on *E. crassipes* leaf diameter (cm)

Treatment	Mean±SE	Treatment	Mean±SE	Treatment	Mean±SE
b0 e0 p1	4.1±0.3 eidhgf	b1 e1 p2	6.9±0.2 ba	b2 e2 p3	5.4±0.5 ebidhagcf
b0 e0 p2	6.3±0.8 ebdac	b1 e1 p3	6.8±1.0 bdac	b2 e3 p1	3.9±0.4 eihgf
b0 e0 p3	6.6±0.7 bdac	b1 e2 p1	4.2±0.3 eidhgef	b2 e3 p2	4.9±0.2 ebidhagcf
b0 e1 p1	4.0±0.3 eidhgf	b1 e2 p2	5.9±0.4 ebdagcf	b2 e3 p3	4.5±0.4 ebidhagcf
b0 e1 p2	6.2±0.6 ebdacf	b1 e2 p3	5.4±0.4 ebidhagcf	b3 e0 p1	3.7±0.6 ihg
b0 e1 p3	7.1±0.5 a	b1 e3 p1	4.1±0.1 eidhgef	b3 e0 p2	5.4±0.8 ebidhagcf
b0 e2 p1	3.8±0.3 eihgf	b1 e3 p2	5.5±0.3 ebidhagcf	b3 e0 p3	5.6±0.7 ebidhagcf
b0 e2 p2	4.5±0.5 ebidhagcf	b1 e3 p3	4.9±0.9 ebidhagcf	b3 e1 p1	4.0±0.3 eidhgf
b0 e2 p3	6.6±0.6 bdac	b2 e0 p1	4.4±0.3 ebidhagcf	b3 e1 p2	4.9±0.4 ebidhagcf
b0 e3 p1	3.8±0.5 ihgf	b2 e0 p2	5.6±0.5 ebidhagcf	b3 e1 p3	5.8±0.4 ebdhagcf
b0 e3 p2	5.8±0.2 ebdhagcf	b2 e0 p3	6.0±1.1 ebdhagcf	b3 e2 p1	3.9±0.2 eihgf
b0 e3 p3	6.0±0.6 ebdagcf	b2 e1 p1	3.5±0.4 ih	b3 e2 p2	5.1±0.2 ebidhagcf
b1 e0 p1	4.2±0.4 ebidhgef	b2 e1 p2	5.0±0.4 ebidhagcf	b3 e2 p3	5.1±0.6 ebidhagcf
b1 e0 p2	6.8±0.2 bac	b2 e1 p3	7.1±0.3 a	b3 e3 p1	3.7±0.2 ihgf
b1 e0 p3	7.2±1.0 a	b2 e2 p1	3.9±0.3 eihgf	b3 e3 p2	5.5±0.1 ebidhagcf
b1 e1 p1	3.4±0.2 i	b2 e2 p2	5.3±0.6 ebidhagcf	b3 e3 p3	4.7±0.8 ebidhagcf

Letters under column "Treatment" refer to weevils (b= *N. bruchi*, e= *N. eichhorniae*) and plants (p= *E. crassipes* plant), while numbers associated with the letters refer to densities (0= control, 1= one pair 2= two pairs and 3= three pairs) and nutrient concentrations (1= low, 2= medium and 3= high) of the weevils and *E. crassipes*, respectively. On columns, SE=standard error. Mean values followed by the same letter(s) indicate no significant difference at the 5% level, Tukey's studentized range test (HSD).

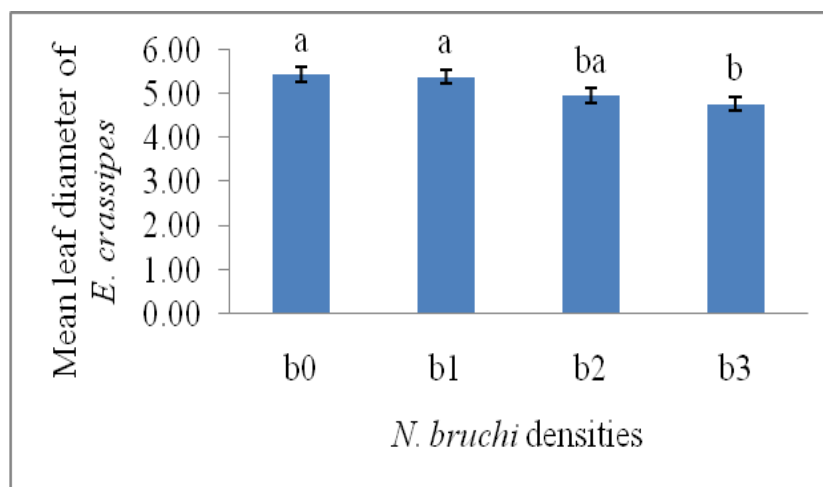


Figure 5. Effect of *N. bruchi* densities, b0-b3 (b= *N. bruchi*, b0= control, b1= one pair, b2= two pairs, and b3= three pairs) on *E. crassipes* leaf diameter (cm). Means sharing the same letter are not significantly different from each other at the 5% level, Tukey's studentized range test (HSD).

Table 7. Effect of nutrient levels, *N. bruchi* and *N. eichhorniae* densities on *E. crassipes* petiole length (cm).

Treatment	Mean± SE	Treatment	Mean± SE	Treatment	Mean± SE
b0 e0 p1	5.53±0.12 ebdcf	b1 e1 p2	7.03±0.38 bdac	b2 e2 p3	6.50±0.51 ebdac
b0 e0 p2	5.80±0.36 ebdacf	b1 e1 p3	5.73±0.24 ebdacf	b2 e3 p1	4.70±0.17 edf
b0 e0 p3	5.80±0.40 ebdacf	b1 e2 p1	5.97±0.29 ebdacf	b2 e3 p2	5.40±0.65 edcf
b0 e1 p1	6.10±0.06 ebdacf	b1 e2 p2	6.70±0.62 bdac	b2 e3 p3	5.47±0.32 ebdcf
b0 e1 p2	5.63±0.19 ebdcf	b1 e2 p3	6.00±0.35 ebdacf	b3 e0 p1	7.00±0.51 bdac
b0 e1 p3	6.60±0.55 bdac	b1 e3 p1	6.57±0.47 ebdac	b3 e0 p2	5.67±0.26 ebdacf
b0 e2 p1	5.00±0.40 edcf	b1 e3 p2	6.20±0.42 ebdac	b3 e0 p3	6.37±0.38 ebdac
b0 e2 p2	5.47±0.24 ebdcf	b1 e3 p3	6.33±0.55 ebdac	b3 e1 p1	4.27±0.44 ef
b0 e2 p3	7.07±0.22 bac	b2 e0 p1	5.53±0.24 ebdcf	b3 e1 p2	5.47±0.41 ebdcf
b0 e3 p1	5.57±0.52 ebdcf	b2 e0 p2	6.27±0.32 ebdac	b3 e1 p3	6.40±0.35 ebdac
b0 e3 p2	6.67±0.44 bdac	b2 e0 p3	6.10±0.67 ebdacf	b3 e2 p1	5.73±0.38 ebdacf
b0 e3 p3	7.73±0.15 ba	b2 e1 p1	3.93±0.37 f	b3 e2 p2	5.03±0.50 edcf
b1 e0 p1	5.07±0.27 edcf	b2 e1 p2	5.67±0.52 ebdacf	b3 e2 p3	5.90±0.50 ebdacf
b1 e0 p2	7.10±0.55 bac	b2 e1 p3	7.90±0.21 a	b3 e3 p1	5.93±0.47 ebdacf
b1 e0 p3	7.00±0.58 bdac	b2 e2 p1	7.20±0.44 bac	b3 e3 p2	5.50±0.25 ebdcf
b1 e1 p1	5.73±0.43 ebdacf	b2 e2 p2	5.27±0.41 edcf	b3 e3 p3	5.03±0.22 edcf

Letters under column "Treatment" refer to weevils (b= *N. bruchi*, e= *N. eichhorniae*) and plants (p= *E. crassipes* plant), while numbers associated with the letters refer to densities (0= control, 1= one pair 2= two pairs and 3= three pairs) and nutrient concentrations (1= low, 2= medium and 3= high) of the weevils and *E. crassipes*, respectively. On columns, SE=standard error. Mean values followed by the same letter(s) indicate no significant difference at the 5% level, Tukey's studentized range test (HSD).

Table 8. Effect of nutrient levels, *N. bruchi* and *N. eichhorniae* densities on *E. crassipes* petiole diameter (cm).

Treatment	Mean± SE	Treatment	Mean± SE	Treatment	Mean± SE
b0 e0 p1	1.93±0.03 ^{fcebdg}	b1 e1 p2	1.13±0.03 ^P	b2 e2 p3	1.50±0.06 ^{nkmjlo}
b0 e0 p2	1.53±0.03 ^{nkmijl}	b1 e1 p3	1.73±0.03 ^{fkeijhg}	b2 e3 p1	2.00±0.06 ^{cebd}
b0 e0 p3	1.23±0.03 ^{P^o}	b1 e2 p1	2.30±0.06 ^a	b2 e3 p2	1.97±0.03 ^{fcebd}
b0 e1 p1	1.13±0.03 ^P	b1 e2 p2	1.50±0.06 ^{nkmjlo}	b2 e3 p3	1.63±0.03 ^{kmijhlg}
b0 e1 p2	1.63±0.03 ^{kmijhlg}	b1 e2 p3	1.53±0.03 ^{nkmjlo}	b3 e0 p1	1.83±0.03 ^{fceihdg}
b0 e1 p3	1.77±0.12 ^{feijhdg}	b1 e3 p1	1.87±0.07 ^{fceibhdg}	b3 e0 p2	1.73±0.03 ^{fkeijhg}
b0 e2 p1	1.37±0.09 ^{nmo}	b1 e3 p2	1.63±0.03 ^{kmijhlg}	b3 e0 p3	1.53±0.03 ^{nkmjlo}
b0 e2 p2	1.93±0.09 ^{fcebdg}	b1 e3 p3	1.43±0.03 ^{nmlo}	b3 e1 p1	1.77±0.03 ^{feijhdg}
b0 e2 p3	2.10±0.17 ^b	b2 e0 p1	1.67±0.03 ^{fkijhlg}	b3 e1 p2	1.47±0.03 ^{nkmlo}
b0 e3 p1	1.30±0.06 ^{npo}	b2 e0 p2	1.87±0.03 ^{fceibhdg}	b3 e1 p3	2.07±0.03 ^{cb}
b0 e3 p2	1.90±0.06 ^{fceibhdg}	b2 e0 p3	1.50±0.06 ^{nkmjlo}	b3 e2 p1	1.50±0.06 ^{nkmjlo}
b0 e3 p3	1.60±0.06 ^{kmijhl}	b2 e1 p1	1.30±0.06 ^{npo}	b3 e2 p2	1.63±0.03 ^{kmijhlg}
b1 e0 p1	1.50±0.06 ^{nkmjlo}	b2 e1 p2	1.60±0.06 ^{kmijhl}	b3 e2 p3	2.00±0.06 ^{cebd}
b1 e0 p2	1.57±0.03 ^{nkmijl}	b2 e1 p3	1.97±0.03 ^{fcebd}	b3 e3 p1	1.63±0.03 ^{kmijhlg}
b1 e0 p3	2.07±0.03 ^{cb}	b2 e2 p1	1.83±0.03 ^{fceihdg}	b3 e3 p2	1.87±0.03 ^{fceibhdg}
b1 e1 p1	1.30±0.06 ^{npo}	b2 e2 p2	1.47±0.03 ^{nkmlo}	b3 e3 p3	2.03±0.07 ^{cbd}

Letters under column "Treatment" refer to weevils (b= *N. bruchi*, e= *N. eichhorniae*) and plants (p= *E. crassipes* plant), while numbers associated with the letters refer to densities (0= control, 1= one pair 2= two pairs and 3= three pairs) and nutrient concentrations (1= low, 2= medium and 3= high) of the weevils and *E. crassipes*, respectively. On columns, SE=standard error. Mean values followed by the same letter(s) indicate no significant difference at the 5% level, Tukey's studentized range test (HSD).

Effect of water nutrient levels, *N. bruchi* and *N. eichhorniae* densities on the number of *Eichhornia crassipes* leaves

There was nonsignificant ($p > 0.05$) interaction between the variables on *E. crassipes* leaf. The number of *E. crassipes* leaves was significantly affected by *N. bruchi* ($p < 0.05$) and water nutrient levels ($p < 0.05$), and it was the highest when it was free of *N. bruchi* and received the highest nutrient level (Figure 6A and B). Except for the treatment with the three pairs of *N. bruchi*, the *E. crassipes* mean leaf number in the untreated control was not significantly different from that in the other treatments (Figure 6A).

Eichhornia crassipes that was exposed to both low water nutrient level and high *N. bruchi* density, the mean leaf number decreased significantly (Figure 6A and B), whereas, *N. eichhorniae* had no significant effect on the *E. crassipes* mean leaf number. However, in another study, *E. crassipes* subjected to a greater mean number of *N. eichhorniae* damaged more quickly and produced fewer leaves (Center and Van, 1989). This shows that low water nutrient level (Yu *et al.*, 2019) and high *N. bruchi* density had a negative impact on *E. crassipes* mean leaf number (Firehun Yirefu *et al.*, 2016).

There was nonsignificant ($p \geq 0.5$) interaction between *N. bruchi*, *N. eichhorniae*, and water nutrient levels on *E. crassipes* leaf loss, but all had a

significant ($p < 0.05$) effect on leaf loss as a single factor (Figure 7 A, B, and C). The mean leaf loss of *E. crassipes* in the highest nutrient level treatments was significantly different from the mean within the untreated control. The mean leaf loss of the *E. crassipes* increased with increasing water nutrient concentrations and decreased with a decrease in the weevil density (Figure 7A, B, and C). Similar to the current study, other studies reported that the *E. crassipes* leaf damage and loss were high as the density of *N. bruchi* and *N. eichhorniae* increased (Kariuki and Minteer, 2021), but *E. crassipes* productivity increased with a decrease in the feeding intensity of the herbivores when they were at a lower density (Bownes *et al.*, 2010). The results imply that the biocontrol of *E. crassipes* is influenced by both herbivore densities and water nutrient levels. Comparing each *N. bruchi* and *N. eichhorniae* independently, *N. bruchi* resulted in a higher number of *E. crassipes* leaf losses and was more effective than *N. eichhorniae* due to its higher fecundity (Bashir *et al.*, 1984; Firehun Yirefu *et al.*, 2016). Although *E. crassipes* density reacts favorably to rising water nutrient levels (Prasetyo *et al.*, 2021), leaf turnover is increased in crowded *E. crassipes*, comparable to when it was infested by herbivores (Center and Van, 1989). This is supported by the current investigation, which indicates that the mean leaf loss trend of *E. crassipes* increased as nutrient levels increased.

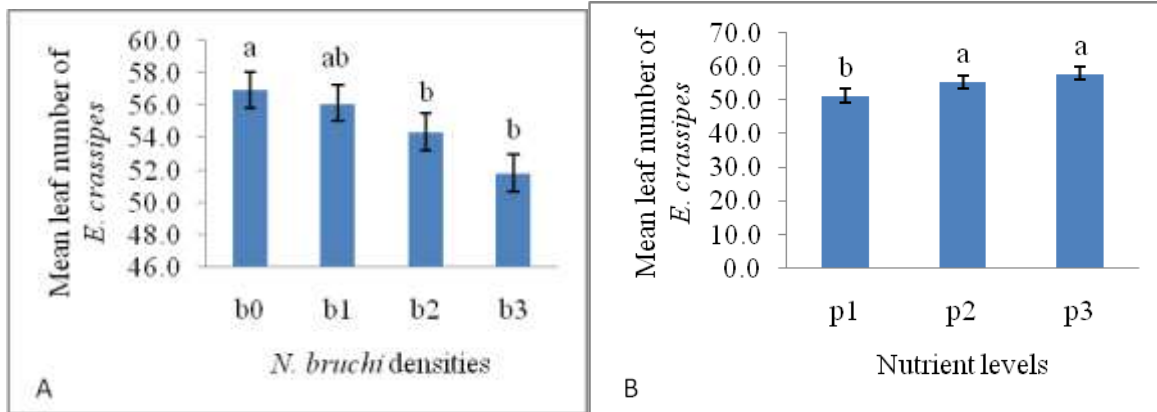


Figure 6. Effect of *N. bruchi* densities, b0-b3 (b= *N. bruchi*, b0= control, b1= one pair, b2= two pairs, and b3= three pairs) on *E. crassipes* leaf number (A) and effect of nutrient levels, p1-p3 (p= *E. crassipes* plant, p1= low, p2= medium, and p3= high) on *E. crassipes* leaf number (B). Means sharing the same letter are not significantly different from each other at the 5% level, Tukey's studentized range test (HSD).

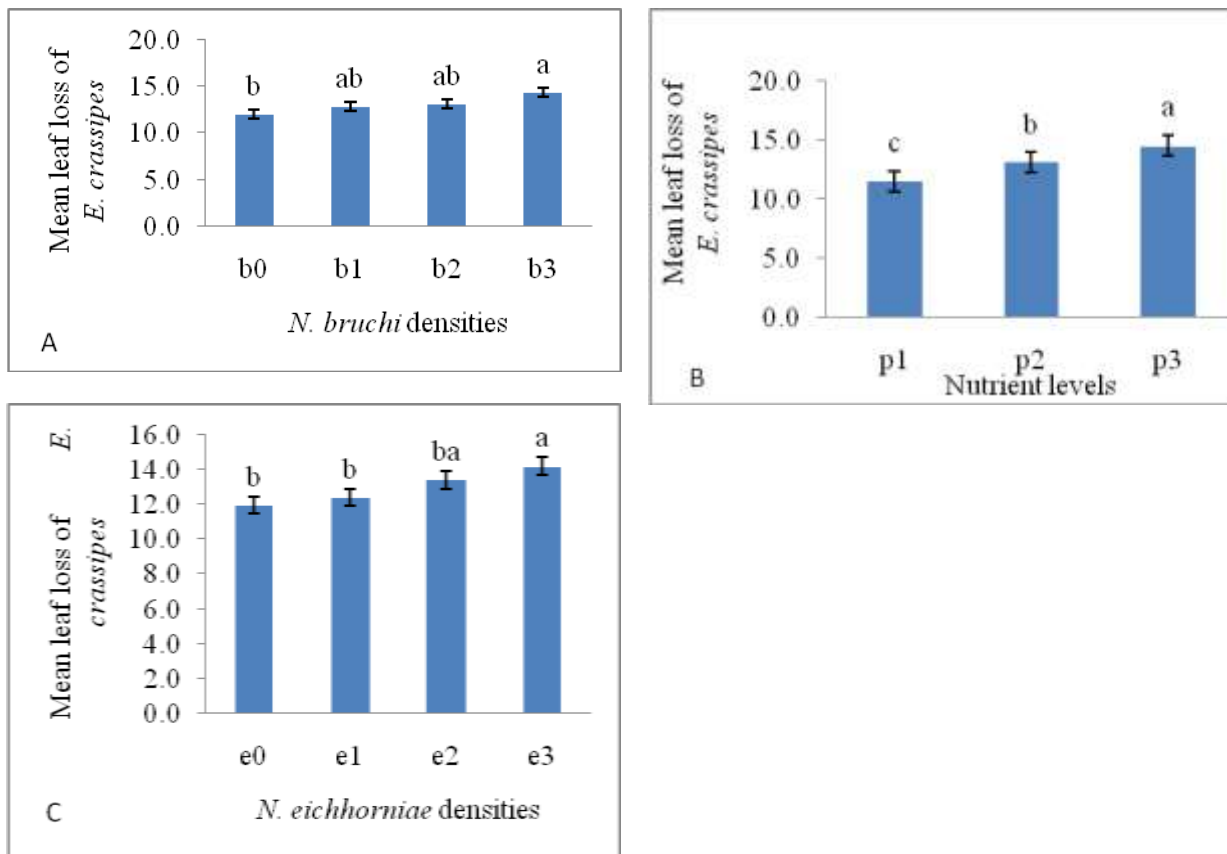


Figure 7. Effect of *N. bruchi* densities, b0-b3 (b= *N. bruchi*, b0= control, b1= one pair, b2= two pairs, and b3= three pairs) on *E. crassipes* leaf loss (A), effect of nutrient levels, p1-p3 (p= *E. crassipes* plant, p1= low, p2= medium, and p3= high) on *E. crassipes* leaf loss (B) and effect of *N. eichhorniae* densities, e0-e3 (e= *N. eichhorniae*, e0= control, e1= one pair, e2= two pairs, and e3= three pairs) on *E. crassipes* leaf loss (C). Means sharing the same letter are not significantly different from each other at the 5% level, Tukey's studentized range test (HSD).

Effect of nutrient levels, *Neochetina bruchi* and *Neochetina eichhorniae* densities on the number of *Eichhornia crassipes* ramets and flowers

The ramet number of the *E. crassipes* was nonsignificantly ($p \geq 0.05$) affected by the interaction of the water nutrient levels, *N. bruchi* and *N. eichhorniae*. The presence of *N. bruchi* and *N. eichhorniae* had nonsignificant ($p > 0.05$) effect on the *E. crassipes* ramet number, but the water nutrient levels did ($p < 0.05$). In an earlier study, Coetzee *et al.* (2007) reported that, herbivore damage did not have as great an effect on *E. crassipes* potential as water nutrient concentrations did. In this study, the *E. crassipes* ramet mean number record was significantly high (5.58 per plot) for the high nutrient level and nearly twofold (1.7) relative to the lowest levels (3.38 per plot) (Figure 8). This is in line with what Yu *et al.* (2019) reported for *E. crassipes* plants at high nutrient level, which could be more than double that in the low nutrient level treatment. This challenge is severe because *E. crassipes* growth and reproduction happen faster than weevils can control them (Ray and Pandey, 2009).

Neochetina eichhorniae had nonsignificant effect on *E. crassipes* flower number either with interaction or as a main effect ($p > 0.05$). The flower number was significantly affected by nutrient levels in *E. crassipes* ($p < 0.05$) and the density of *N. bruchi* ($p < 0.05$). The mean *E. crassipes* flower number was the highest in the untreated control and significantly different from that in the other treatments. It showed a decreasing trend with an increase in *N. bruchi* density and water nutrient levels (Figure 9A and B). This reduction in inflorescence was related to damage caused by *N. bruchi*, which feeds on *E. crassipes* and subsequently affects seed production (Fayad *et al.*, 2008; Gupta and Yadav, 2020). The current study found that the highest nutrient level treatments reduced the mean number of *E. crassipes* flowers. This was due to the high nutrient treatment level which exhibited a better relative growth rate than the low nutrient treatment level, which had a higher allocation to sexual reproduction (Soti and Volin, 2010). Water nutrient enrichment, such as nitrogen, increases the rate of asexual reproduction and leaf turnover in *E. crassipes* (Wilson *et al.*, 2005).

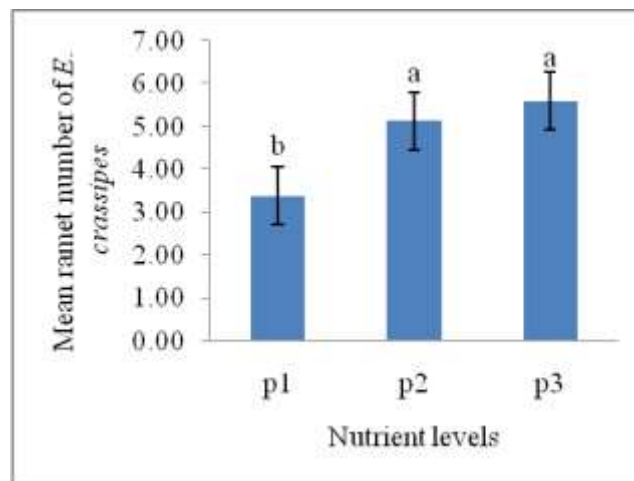


Figure 8. Effect of nutrient levels, p1-p3 (p= *E. crassipes* plant, p1= low, p2= medium, and p3= high) on *E. crassipes* ramet number. Means sharing the same letter are not significantly different from each other at the 5% level, Tukey's studentized range test (HSD).

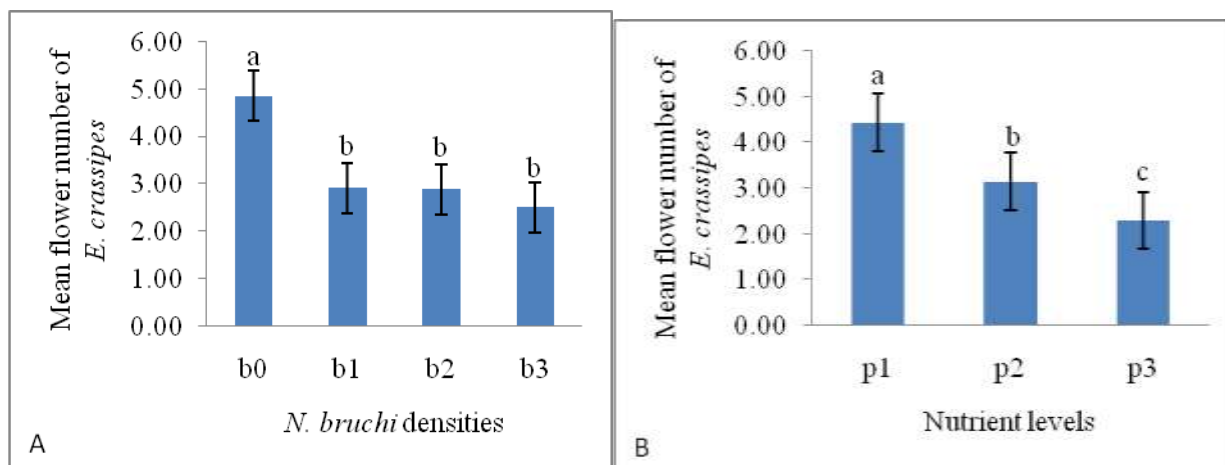


Figure 9. Effect of *N. bruchi* densities, b0-b3 (b= *N. bruchi*, b0= control, b1= one pair, b2= two pairs, and b3= three pairs) on *E. crassipes* flower number (A) and effect of nutrient levels, p1-p3 (p= *E. crassipes* plant, p1= low, p2= medium, and p3= high) on *E. crassipes* flower number (B). Means sharing the same letter are not significantly different from each other at the 5% level, Tukey’s studentized range test (HSD).

Association between *Eichhornia crassipes* growth parameters

Leaf number had a significant positive correlation with fresh weight ($r = 0.58, p < 0.05$), dry weight ($r = 0.36, p < 0.05$), leaf length ($r = 0.32, p < 0.05$), leaf diameter ($r = 0.23, p < 0.05$), petiole

length ($r = 0.26, p < 0.05$), leaf loss ($r = 0.28, p < 0.05$), and ramet number ($r = 0.59, p < 0.05$) (Table 9). However, leaf number had a nonsignificant negative correlation with petiole diameter ($r = -0.03, p > 0.05$) and flower number ($r = -0.16, p \geq 0.05$) (Table 9).

Table 9. Correlation coefficient of *E. crassipes* growth parameters.

	Fresh	Dry	Lfl	Lfdm	Ptl	Ptldm	Lfno	Lfloss	Rmtno	Flrno
Fresh	1									
Dry	.401**	1								
Lfl	.357**	.309**	1							
Lfdm	.242**	.301**	.882**	1						
Ptl	.249**	.094	.443**	.383**	1					
Ptldm	-.041	-.092	.138	.119	.102	1				
Lfno	.584**	.355**	.322**	.234**	.264**	-.031	1			
Lfloss	-.066	-.134	-.018	.066	.039	.060	.284**	1		
Rmtno	.513**	.316**	.409**	.441**	.291**	.056	.587**	.187*	1	
Flrno	.050	.016	-.103	-.204*	-.183*	-.133	-.162	-.320**	-.320**	1
	.554	.850	.219	.014	.028	.111	.052	.000	.000	.000

** = Correlation is significant at the 0.01 level (2-tailed), * = Correlation is significant at the 0.05 level (2-tailed). Lfl = leaf length, Lfdm = leaf diameter, Ptl = petiole length, Ptldm = petiole diameter, Lfno = leaf number, Lfloss = leaf loss, Rmtno = ramet number, Flrno = flower number.

CONCLUSION

The present study indicates that, changes in *E. crassipes* quality affected a feeding potential and population dynamics of *N. bruchi* and *N. eichhorniae* as they feed on it. Density of the weevils and their feeding potential increased on *E. crassipes* under eutrophic nutrient conditions. Based on these findings, more weevils should be released into heavily infested *E. crassipes* to provide effective and faster control by compensating the weevils' delayed life cycle. Because of their different responses to *E. crassipes* under varied nutrient concentrations, the combined release of the two weevils must be employed for better management. Field release and postrelease evaluation of *N. bruchi* and *N. eichhorniae* need to be considered. In integrated *E. crassipes* management, the effective use of weevils should be emphasized to address the enormous socioeconomic, ecological, industrial, and domestic concerns that Ethiopia and the rest of the world face.

ACKNOWLEDGMENT

We would like to thank the Bio and Emerging Technology Institute for funding this research activity. Likewise, we acknowledge the Ministry of Education and Addis Ababa University for additional budget and facilities for the success of this research work. We are also grateful to Ethiopian Sugar Corporation, Research and Development Center and its entire staff for their technical and other cooperation in this study.

REFERENCES

- Addis Anteneh and Hailu Yemanu (1970). Development of the Awash Valley. *Zede Journal* **4**:19-24.
- Adugnaw Admas, Samuuel Sahle, Erehmet Belete, Aklilu Agidie and Mehari Alebachew (2017). Controlling water hyacinth in Lake Tana using biological method at green house and pond level. *European Journal of Experimental Biology* **7**:1-5.
- Ajuonu, O., Schade, V., Veltman, B., Sedjro, K. and Neuenschwander, P. (2003). Impact of the exotic weevils *Neochetina* spp.: (Coleoptera: Curculionidae) on water hyacinth, *Eichhornia crassipes* (Lil: Pontederiaceae) in Benin, West Africa. *African Entomology* **11**:153-161.
- Andualem Mekonnen, Tsigereda Assefa, Kuribel Tesfaye and Derbu Getahun (2014). Spatial distribution of nitrate in the drinking water sources found in Ethiopia; retrospective study. *Global Journal of Environmental Science and Technology* **2**:75-81.
- Bashir, M. O., El Abjar, Z. E. and Irving, N. S. (1984). Observations on the effect of the weevils, *Neochetina eichhorniae* Warner and *Neochetina bruchi* Hustache, on the growth of water hyacinth. *Hydrobiologia* **110**:95-98.
- Bick, E., de Lange, E. S., Kron, C. R., da Silva Soler, L., Liu, J. and Nguyen, H. D. (2020). Effects of salinity and nutrients on water hyacinth and its biological control agent, *Neochetina bruchi*. *Hydrobiologia* **847**:3213-3224.
- Bock, J. H. (1969). Productivity of the water hyacinth *Eichhornia crassipes* (Mart.) Solms. *Ecology* **50**:460-464.
- Bownes, A., Hill, M.P. and Byrne, M.J. (2010). Assessing density-damage relationships between water hyacinth and its grasshopper herbivore. *Entomologia Experimentalis et Applicata* **137**:246-254.
- Bownes, A. (2008). Evaluation of a plant-herbivore system in determining potential efficacy of a candidate biological control agent, *Cornops aquaticum* for water hyacinth, *Eichhornia crassipes*. PhD Dissertation, Rhodes University, South Africa.
- Buchanan, A. L. (2013). Damage by *Neochetina* weevils (Coleoptera: Curculionidae) induces resistance in *Eichhornia crassipes* (Commelinales: Pontederiaceae). *Florida Entomologist* **96**:458-462.
- Center, T.D. and Dray Jr, F.A. (1992). Associations between water hyacinth weevil (*Neochetina eichhorniae* and *N. bruchi*) and phenological stages of *Eichhornia crassipes* in Southern Florida. *Florida Entomologist* **75**:196-211.
- Center, T. D. and Dray Jr, F. A. (2010). Bottom-up control of water hyacinth weevil populations: do the plants regulate the insects? *Journal of Applied Ecology* **47**:329-337.
- Center, T. D., Steward, K. K. and Bruner, M. C. (1982). Control of waterhyacinth (*Eichhornia crassipes*) with *Neochetina eichhorniae* (Coleoptera: Curculionidae) and a growth retardant. *Weed Science* **30**:453-457.
- Center, T. D. and Van, T. K. (1989). Alteration of water hyacinth (*Eichhornia crassipes* (Mart.) Solms) leaf dynamics and phytochemistry by insect damage and plant density. *Aquatic Botany* **35**:181-195.
- Cilliers, C. J., Hill, M. P., Ogwang, J. A. and Ajuonu, O. (2003). Aquatic weeds in Africa and their

- control. In: Biological Control in IPM Systems in Africa, pp. 161–178 (Neuenschwander, P., Borgemeister, C. and Langewald, J., eds.). CAB International, London.
16. Coetzee, J. A., Byrne, M. J. and Hill, M. P. (2007). Impact of nutrients and herbivory by *Eccritotarsus catarinensis* on the biological control of water hyacinth, *Eichhornia crassipes*. *Aquatic Botany* **86**:179–186.
 17. Coetzee, J. A. and Hill, M. P. (2012). The role of eutrophication in the biological control of water hyacinth, *Eichhornia crassipes*, in South Africa. *Biological Control* **57**:247–261.
 18. Dereje Tewabe (2015). Preliminary survey of water hyacinth in Lake Tana, Ethiopia. *Global Journal of Allergy* **1**:13–18.
 19. Delfosse, E. S., Sutton, D. L. and Perkins, B. D. (1976). Combination of the mottled waterhyacinth weevil and the white amur for biological control of waterhyacinth. *Journal of Aquatic Plant Management* **14**:64–67.
 20. DeLoach, C. J. (1976). *Neochetina bruchi*, a biological control agent of water hyacinth: host specificity in Argentina. *Annals of the Entomological Society of America* **69**:635–642.
 21. DeLoach, C. J. and Cordo, H. A. (1976). Life cycle and biology of *Neochetina bruchi*, a weevil attacking water hyacinth in Argentina, with notes on *N. eichhorniae*. *Annals of the Entomological Society of America* **69**:643–652.
 22. Dissanayake, D., Ranaweera, B. and Amarasingha, A. A. L. (2009). Effect of sex ratio in *Neochetina bruchi* adult population on their performance for biological control of *Eichhornia crassipes*. In: Proceedings of 9th Agricultural Research Symposium, pp. 372, Wayamba University, Sri Lanka.
 23. Fayad, Y. H., Shalaby, F. F., Hafez, A. A. and El-Zoghby, I. R. M. (2008). Impact of applying *Neochetina bruchi* (Mustache), (Coleoptera: Curculionidae) on flowering and water loss through plant leaves for biological control of water hyacinth. *Egyptian Journal of Biological Pest Control* **18**:221–225.
 24. Fekadu Aduna, Fekadu Fufa, Tamene Adugna and Januszkiewicz, K. (2021). Hydroclimate trend analysis of Upper Awash Basin, Ethiopia. *Water* **13**, 1680.
 25. Firehun Yirefu, Struik, P. C., Lantinga, E. A. and Taye Tessema (2014). Water hyacinth in the Rift Valley water bodies of Ethiopia: its distribution, socio-economic importance and management. *International Journal of Current Agricultural Research* **3**:67–75.
 26. Firehun Yirefu, Struik, P. C., Lantinga, E. A. and Taye Tessema (2015). Adaptability of two weevils (*Neochetina bruchi* and *Neochetina eichhorniae*) with potential to control water hyacinth in the Rift Valley of Ethiopia. *Crop Protection* **76**:75–82.
 27. Firehun Yirefu, Struik, P. C., Lantinga, E. A. and Taye Tessema (2016). Pre-release evaluation of *Neochetina* weevils potential for the management of *Eichhornia crassipes* [Mart.] Solm. In the Rift Valley of Ethiopia. *Academia Journal of Agricultural Research* **4**:394–403.
 28. Gopal, B. (1987). Aquatic plant studies1: Water hyacinth. Netherlands: Elsevier Science.
 29. Gore, P. G. (2017). Management of water hyacinth, *Eichhornia crassipes* (Mart.) Solms through biocontrol agents with special reference to *Neochetina* spp. At Raipur District (CG). PhD Thesis, Indira Gandhi Krishi Vishwavidhyalaya, Raipur.
 30. Gore, P., Ganguli, J. and Gauraha, R. (2020). Comparative feeding efficiency of *Neochetina bruchi* and *N. eichhorniae* on water hyacinth under laboratory conditions at Raipur, Chhattisgarh. *Journal of Entomology and Zoology Studies* **8**:1226–1231.
 31. Goyer, R.A. and Stark, J.D. (1984). The impact of *Neochetina eichhorniae* on water hyacinth in southern Louisiana. *Journal of Aquatic Plant Management* **22**:57–61.
 32. Gupta, A. K. and Yadav, D. (2020). Biological control of water hyacinth. *Environmental Contaminants Reviews* **3**:37–39.
 33. Harley, K.L.S., Julien, M.H. and Wright, A.D. (1996). Water hyacinth: a tropical worldwide problem and methods for its control. In: Proceedings of the Second International Weed Control Congress, pp. 639–644 (Brown, H., Cussans, G.W., Devine, M.D., Duke, S.O., Fernandez-Quintanilla, C., Helweg, A., Labrada, R.E., Landes, M., Kudsk, P. and Streibig, J.C., eds). Copenhagen (DK).
 34. Heard, T. A. and Winterton, S. L. (2000). Interactions between nutrient status and weevil herbivory in the biological control of water hyacinth. *Journal of Applied Ecology* **37**:117–127.
 35. Hopper, J. V., Pratt, P. D., McCue, K. F., Pitcairn, M. J., Moran, P. J. and Madsen, J. D. (2017). Spatial and temporal variation of biological control agents associated with *Eichhornia crassipes* in the Sacramento-San Joaquin River Delta, California. *Biological Control* **111**:13–22.
 36. Hopper, J. V., Pratt, P. D., Reddy, A. M., McCue, K. F., Rivas, S. O. and Grosholz, E. D. (2021). Abiotic and biotic influences on the performance of two biological control agents, *Neochetina bruchi* and *N. eichhorniae*, in the

- Sacramento-San Joaquin River Delta, California (USA). *Biological Control* 153,104495.
37. Ismail, M., Compton, S. G. and Brooks, M. (2017). Interaction between temperature and water nutrient levels on the fitness of *Eccritotarsus catarinensis* (Hemiptera: Miridae), a biological control agent of water hyacinth. *Biological Control* 106:83–88.
 38. Jadhav, A., Hill, M. and Byrne, M. (2008). Identification of a retardant dose of glyphosate with potential for integrated control of water hyacinth, *Eichhornia crassipes* (Mart.) Solms-Laubach. *Biological Control* 47:154–158.
 39. Jones, R. W., Hill, J. M., Coetzee, J. A. and Hill, M. P. (2018). The contributions of biological control to reduced plant size and biomass of water hyacinth populations. *Hydrobiologia* 807:377–388.
 40. Julien, M.H., Griffiths, M.W. and Wright, A.D. (1999). Biological Control of Water Hyacinth: the Weevils *Neochetina bruchi* and *N. eichhorniae*: Biologies, Host Ranges, and Rearing, Releasing and Monitoring Techniques for Biological Control of *Eichhorniae crassipes*. ACIAR Monograph No. 60.
 41. Julien, M. H. (2000). Biological control of water hyacinth with arthropods: a review to 2000. In: Aciar Proceedings, pp. 8–20, Queensland, Australia.
 42. Julien, M.H. (2001). Biological control of water hyacinth with arthropods: a review to biological and integrated control of water hyacinth, *Eichhornia crassipes*. In: Proceedings of the Second Meeting of the Global Working Group for the Biological and Integrated Control of Water Hyacinth, pp. 8–20 (Julien, M.H., Hill, M.P., Center, T.D. and Jianqing, D., eds). Queensland, Australia.
 43. Kariuki, E. and Minter, C. (2021). Mottled Water Hyacinth Weevil *Neochetina eichhorniae* Warner (Insecta: Coleoptera: Curculionidae). EENY-741, Electronic Document Information System (EDIS 2021, 5–5). URL <https://edis.ifas.ufl.edu>.
 44. Kidd, H. (2000). Water hyacinth control-an update. *Pesticide Outlook* 11:107–108.
 45. Little, E. C. S. (1965). The world wide distribution of the water hyacinth. *Hyacinth Control Journal* 4:30–32.
 46. LVEMP (2006). Implementation Completion Report on Two International Development Association Credits and a Global Environment Facility Grant in the Amount of US\$ 29.74 Million to the Government of Uganda for the Lake Victoria Environmental Management Project. Report No. 36559.
 47. LVEMP (2016). Climate Change, Pollution and Sustainable Use of Natural Resources in the Lake Victoria Region. Current Situation of Lake Victoria – Facts, Figures and Experience. Anna Masasi, ES – LVEMP II Report.
 48. Mailu, A. M. (2001). Preliminary assessment of the social, economic and environmental impacts of Water Hyacinth in Lake Victoria basin and status of control. In: Proceedings of the Second Meeting of the Global Working Group for the Biological and Integrated Control of Water Hyacinth, pp. 130–139, Beijing, China.
 49. Manning, J. H. (1979). Establishment of waterhyacinth weevil populations in Louisiana. *Journal of Aquatic Plant Management* 17:39–41.
 50. Moran, P. J. (2006). Water nutrients, plant nutrients, and indicators of biological control on waterhyacinth at Texas field sites. *Journal of Aquatic Plant Management* 44:109–114.
 51. Mukarugwiro, J. d' A., Newete, S. W., Hauptfleisch, K. and Byrne, M. J. (2018). The effect of water nutrients on the feeding intensity and development of larvae of *Neochetina eichhorniae* (Warner)(Coleoptera: Curculionidae), a biocontrol agent of the invasive water hyacinth. *African Entomology* 26:63–72.
 52. Mumma, A.C. (1999). The Lake Victoria water hyacinth: its implications for international environmental conflicts management and regional relations in East Africa. MSc Thesis in International Studies, at the Institute of Diplomacy and International Studies, University of Nairobi, Nairobi, Kenya.
 53. Nagassa Dechassa and Belay Abate (2020). Current status of water hyacinth (*Eichhornia crassipes*) in Ethiopia: achievements, challenges and prospects: a review. *Journal of Environment and Earth Science* 10:36–47.
 54. Njoka, S.W. (2004). The biology and impact of *Neochetina* weevils on water hyacinth, *Eichhornia crassipes*, in Lake Victoria Basin. PhD Dissertation, Moi University, Kenya.
 55. O'Brien, C. W. (1976). A taxonomic revision of the new world subaquatic genus *Neochetina* (Coleoptera: Curculionidae: Bagoini). *Annals of the Entomological Society of America* 69:165–174.
 56. Prasetyo, S., Anggoro, S. and Soeprbowati, T. R. (2021). The Growth rate of water hyacinth (*Eichhornia crassipes* (Mart.) Solms) in Rawapening Lake, Central Java. *Journal of Ecological Engineering* 22:222–231.
 57. Pratiwi, F. D., Zainuri, M., Purnomo, P. W. and Purwanti, F. (2018). Evaluation of weevils (*Neochetina* spp.) existence in Rawa Pening Lake (abundance and impact on target and non-target plant). *Fluxion Biosciences* 11:823–832.

58. Ray, P. (2015). Population dynamics and sex ratio of two biocontrol agents of water hyacinth. *Indian Journal of weed Science* **47**:188-192.
59. Ray, P. and Pandey, A. K. (2009). Impact evaluation of *Neochetina* spp. On different growth stages of waterhyacinth. *Journal of Plant Protection Research* **48**:8-13.
60. Reddy, K. R., Agami, M. and Tucker, J. C. (1989). Influence of nitrogen supply rates on growth and nutrient storage by water hyacinth (*Eichhornia crassipes*) plants. *Aquatic Botany* **36**:33-43.
61. Sivaraman, K. and Murugesan, A. G. (2017). Impact of release of *Neochetina* spp. On growth and density of water hyacinth *Eichhornia crassipes*. *Journal of Biological Control* **30**:158-163.
62. Soti, P. G. and Volin, J. C. (2010). Does water hyacinth (*Eichhornia crassipes*) compensate for simulated defoliation? Implications for effective biocontrol. *Biological Control* **54**:35-40.
63. Voukeng, S. N. K. (2017). Biotic and abiotic factors promoting the development and proliferation of water hyacinth (*Eichhornia crassipes* (Mart.) Solms-Laub.) in the Wouri Basin (Douala-Cameroon) and environs, with implications for its control. PhD Thesis, Rhodes University, South Africa.
64. Wassie Anteneh, Dereje Tewabe, Addisalem Assefa, Abebaw Zeleke, Befta Tenaw and Yitayew Wassie (2015). Water hyacinth coverage survey report on Lake Tana Biosphere Reserve. In: Technical Report Series 2, pp. 1-25, Bahir Dar University, Ethiopia.
65. Wilson, J. R., Holst, N. and Rees, M. (2005). Determinants and patterns of population growth in water hyacinth. *Aquatic Botany* **81**:51-67.
66. Wilson, J. R. U., Rees, M. and Ajuonu, O. (2006). Population regulation of a classical biological control agent larval density dependence in *Neochetina eichhorniae* Coleoptera Curculionidae, a biological control agent of water hyacinth *Eichhornia crassipes*. *Bulletin of Entomological Research* **96**:145-152.
67. Wise, R. M., Van Wilgen, B. W., Hill, M. P., Schulthess, F., Tweddle, D., Chabi-Olay, A. and Zimmermann, H. G. (2007). The economic impact and appropriate management of selected invasive alien species on the African continent. *Global Invasive Species Programme*, **19**:282-295.
68. Wright, A. D. and Center, T. D. (1984). Predicting population intensity of adult *Neochetina eichhorniae* (Coleoptera: Curculionidae) from incidence of feeding on leaves of waterhyacinth, *Eichhornia crassipes*. *Environmental Entomology* **13**:1478-1482.
69. Yirga Kebede (2016). Water quality status of Lake Tana, Ethiopia. *Journal of Civil and Environmental Research* **8**:39-41.
70. Yu, H., Dong, X., Yu, D., Liu, C. and Fan, S. (2019). Effects of eutrophication and different water levels on overwintering of *Eichhornia crassipes* at the northern margin of its distribution in China. *Frontiers in Plant Science* **10**:1-10.