Short communications

A quantitative post-release evaluation of biological control of water lettuce, *Pistia stratiotes* L. (Araceae) by the weevil *Neohydronomus affinis* Hustache (Coleoptera: Curculionidae) at Cape Recife Nature Reserve, Eastern Cape Province, South Africa

G.R. Moore & M.P. Hill*

Department of Zoology and Entomology, Rhodes University, P.O. Box 94, Grahamstown, 6140 South Africa

Water lettuce, *Pistia stratiotes* L. (Araceae) is recognized as being among the world's worst aquatic weeds. In its adventive range, the plant forms extensive mats capable of blocking navigation channels, impeding water flow in irrigation and flood control canals, and disrupting hydropower generation (Holm *et al.* 1977). Dense mats of the weed prevent light penetration into the water column which negatively affects submerged aquatic plant communities, causing a lowering of the oxygen concentration and thereby reducing benthic invertebrate and fish populations (Neuenschwander *et al.* 2009).

Biological control of water lettuce has been highly successful in most areas around the world through the introduction of the weevil, *Neohydronomus affinis* Hustache (Coleoptera: Curculionidae) (Neuenschwander *et al.* 2009). This agent has been successfully released in at least 10 countries: Australia, Benin, Botswana, Ghana, Papua New Guinea, South Africa, Senegal, United States of America, Zambia and Zimbabwe (Julien & Griffiths 1998). However, information on the impact of the agent on weed populations has largely been anecdotal and seldom quantified.

Adult *N. affinis* are small (3 mm long) are bluish grey to brown (depending on the age) with dense scales on the elytra. The eggs are cream and spherical (0.3 mm × 0.4 mm). Females chew a hole in the water lettuce leaf (usually on the upper surface), deposit a single egg and close the hole with frass. The eggs hatch within about four days at temperatures above 24 °C (Center *et al.* 2002). The insect goes through three larval instars that take between 11 and 14 days during which time they mine the leaves, causing severe damage to the plants, and they then pupate in the leaves. Adult feeding causes small shot-holes in the younger leaves. Generation time, depending on temperature, is 4–6 weeks (Neuenschwander *et al.* 2009).

Neohydronomus affinis was first introduced into South Africa in 1985 onto a water lettuce infestation on Nhlangaluwe Pan in the Kruger National Park (Cilliers 1987). Since then the weevil has been released on, or has spread to most water lettuce infestations in the country (Coetzee *et al.* 2011). The control of water lettuce has been highly successful in South Africa (Cilliers 1991) as it has in most parts of the world (Neuenschwander *et al.* 2009), and in many areas the weed is no longer considered problematic.

Although biological control is considered the most sustainable method for reducing populations of this weed to below an economic and/or ecological threshold, in small, eutrophic water bodies in temperate areas, biological control has been less successful (Hill 2003). In these systems, although insect populations are high, the plants are able to tolerate herbivory through rapid leaf turnover facilitated by high levels of nitrates and phosphates (Ripley *et al.* 2006).

Biological control of floating aquatic weeds is traditionally less successful where the plants are growing in conditions of high nutrients in that the weed populations are not reduced below the acceptable level of control (Room & Thomas 1985; Coetzee & Hill 2012), or as was the case with water lettuce control on Sunset Dam in the Kruger National Park, the time taken to achieve control is longer (five to seven years) than in oligothrophic systems (one to two years) (Cilliers 1991).

Water lettuce was first noticed on the wastewater treatment settlement pond at the Cape Recife Nature Reserve, Port Elizabeth, Eastern Cape Province (34°01′11.9″S 25°41′18.7″E) in March 2002, and within two months it had completely

*To whom correspondence should be addressed.
E-mail: m.p.hill@ru.ac.za

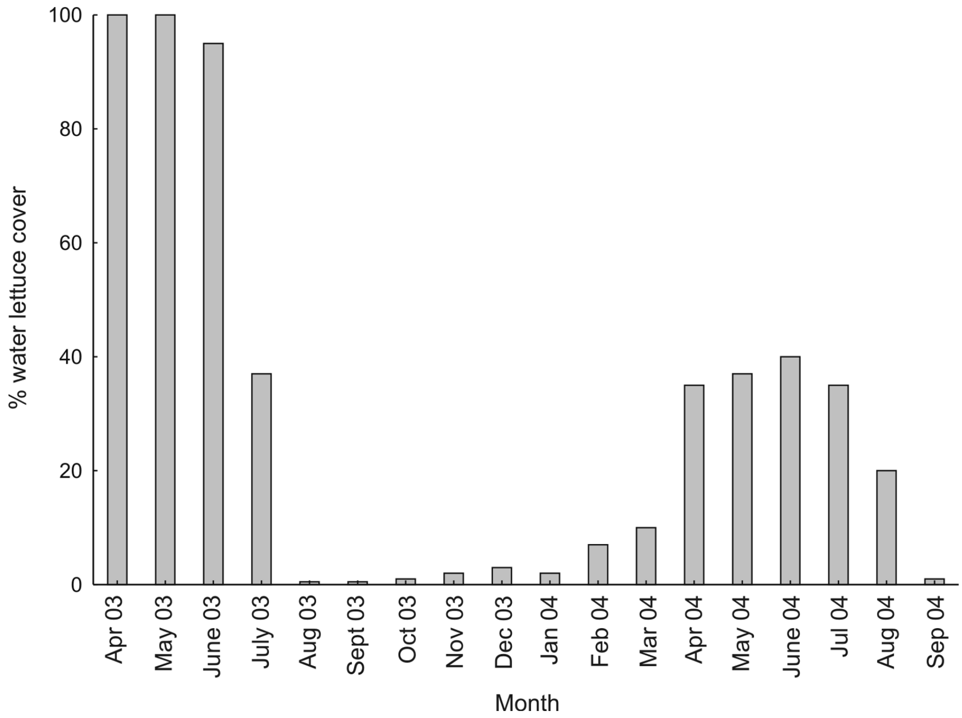

Fig. 1. Percentage coverage of *Pistia stratiotes* on the sewerage settlement pond at Cape Recife from April 2003 (seven months after the introduction of the weevils) to September 2004.

covered the 1.5-ha pond. In August 2002, 240 adult *N. affinis,* which were sourced from the Rhodes University mass-rearing facility in Grahamstown, were released on the pond and a post-release evaluation project was initiated in May 2003. Quantitative data were collected monthly for a year. Long-term post-release evaluations in weed biological control are important definitive proof of the success of any project and inform weed managers whether or not to research and release additional agents or implement other control measures. Therefore the aim of this study was to determine the impact of *N. affinis* on water lettuce under highly eutrophic field conditions.

Water quality data (NH_3-nitrogen and NO_3-nitrogen) were received from the Nelson Mandela Metro database for 2003 and 2004, for the ponds at Cape Recife. Water lettuce was sampled monthly on the pond between May 2003 and May 2004. During each sampling event, percentage cover of the dam was estimated and at least 100 water lettuce plants were collected, with the exception of April 2004 when only 34 plants could be collected due to a reduction in plant number caused by the weevils. Plant parameters measured included wet weight and the number of daughter plants (ramets). The number of fruit and seeds per plant were also counted. The number of adult weevils per plant was also counted. A Kolmogorov-Smirnov (K-S) test conducted on all variables, showed that the data were not normally distributed and thus the changes in wet weight of plants, number of daughter plants, number of fruits and seeds per plant and the number of weevils per plant were compared over the 12-month sampling period using a Kruskal-Wallis test (Fowler *et al.* 2005).

The pond was highly nutrient enriched, or eutrophic. The mean concentration of ammonia (NH_3) in the pond for 187 samples taken over a period of two years (2003 and 2004), was 8.3 mg/l (±7.3 mg/l, min. 0.1 mg/l, max. 57 mg/l), (n = 275). Mean nitrate (NO_3) levels were 2.0 mg/l (±2.4 mg/l, min. –0.4 mg/l, max. 12 mg/l), (n = 275). The mean value of 8.3 mg/l NH_3 is well within the eutrophic classification of the South African water trophic states, as published in the South African Water Quality Guidelines set by the Department of Water Affairs and Forestry (DWAF) 1996 (De Villiers & Thiart 2007). However, the pond frequently reached nutrient levels well above the minimum

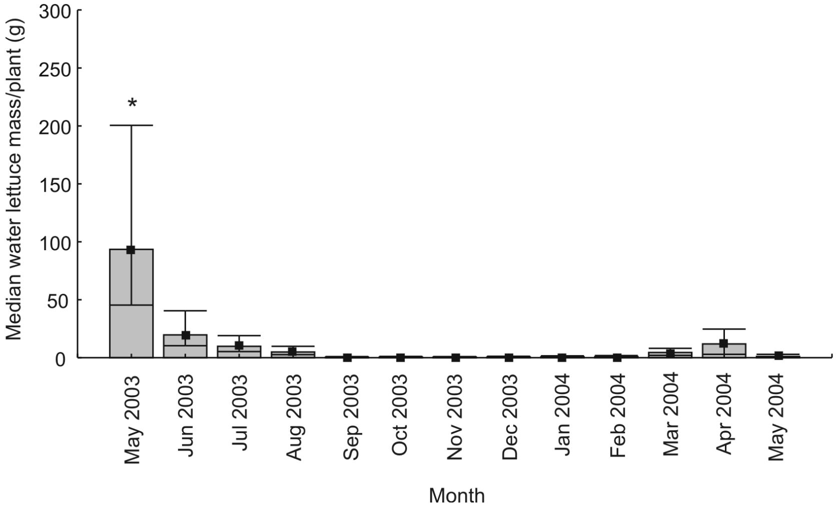

Fig. 2. Median mass of water *Pistia stratiotes* on the sewerage settlement pond at Cape Recife monthly from May 2003 to May 2004 (n = 100 plants). Error bars represent 25 percentiles. *Denotes that the mass of plants at this sampling event was significantly different from the other sampling events ($P < 0.0001$).

hypertrophic value of 10 mg/l NH_3.

The percentage cover of water lettuce at Cape Recife decreased from 100 % in May 2003 to less than 1 % by August 2003, and remained relatively clear of weed over most of summer, increasing again towards the end of summer and into early autumn (Fig. 1). The resurgence of water lettuce towards the end of summer in 2004 is a natural cycle in that the rapid decline in weed coverage by August 2003 would have resulted in considerable larval and pupal mortality and the adults would have dispersed in search of other water lettuce infestations. The few remaining plants would therefore have been able to multiply in the absence of the weevil and there would have been some seedling recruitment. There was a significant decrease in plant wet weight between May 2003 and May 2004 ($H_{(12;\ 2360)} = 2020.34$, $P < 0.0001$) (Fig. 2). At the start of the study, a number of *P. stratiotes* plants were well over 1 kg, but three months into the sampling programme, plants seldom weighed more than 10 g. Ramet produc-

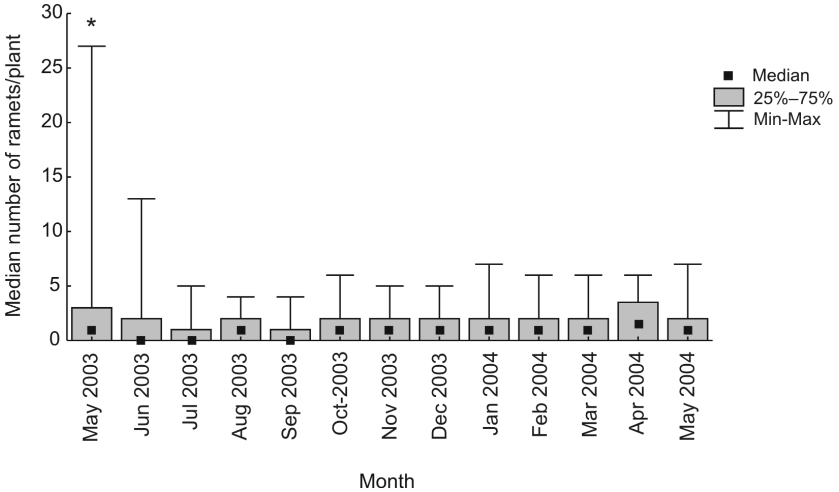

Fig. 3. Median number of *Pistia stratiotes* ramets per plant per month over 12 months of sampling on the sewerage settlement pond at Cape Recife (n = 100 mother plants). *Denotes that the mean number of ramets per plant at this sampling event was significantly higher than the other sampling events ($P < 0.0001$).

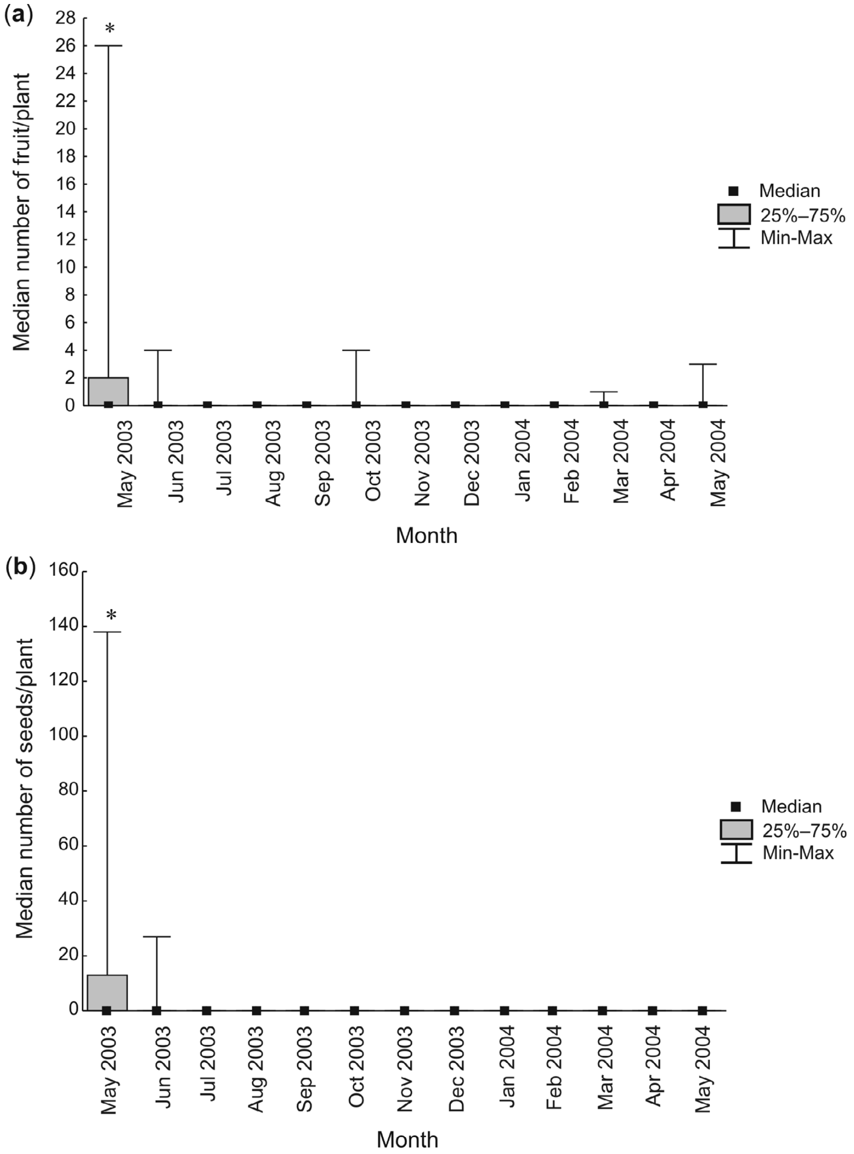

Fig. 4. Median number of fruit (**a**) and seeds (**b**) per plant per month for *Pistia stratiotes* plants sampled at Cape Recife over a 12-month period ($n = 100$ plants). *Denotes a significant difference between this sampling event and the other sampling events ($P < 0.0001$).

tion also decreased significantly over time ($H_{(12;\ 3786)} = 101.57$, $P < 0.0001$) (Fig. 3). There were also significant decreases in the number of fruit ($H_{(12;\ 3786)} = 1248$, $P < 0.0001$) (Fig. 4a) and seeds per plant ($H_{(12;\ 3786)} = 1065.5$, $P < 0.0001$)) (Fig. 4b).

Numbers of weevils at Cape Recife followed a similar trend to that of the plants. In May 2003, eight months after the weevils were released, there was a median number of three larvae per plant and maximum of 25 and a median number of 17 adult weevils per plant and a maximum of 193 adults on one plant. Larval ($H_{(12;\ 3786)} = 722.8$, $P < 0.0001$) (Fig. 5a) and adult weevil numbers ($H_{(12;\ 3786)} = 2403.3$, $P < 0.0001$) (Fig. 5b) per plant decreased significantly over time as plant size decreased.

Although the post-release data presented here only represent a 12-month period, the site has

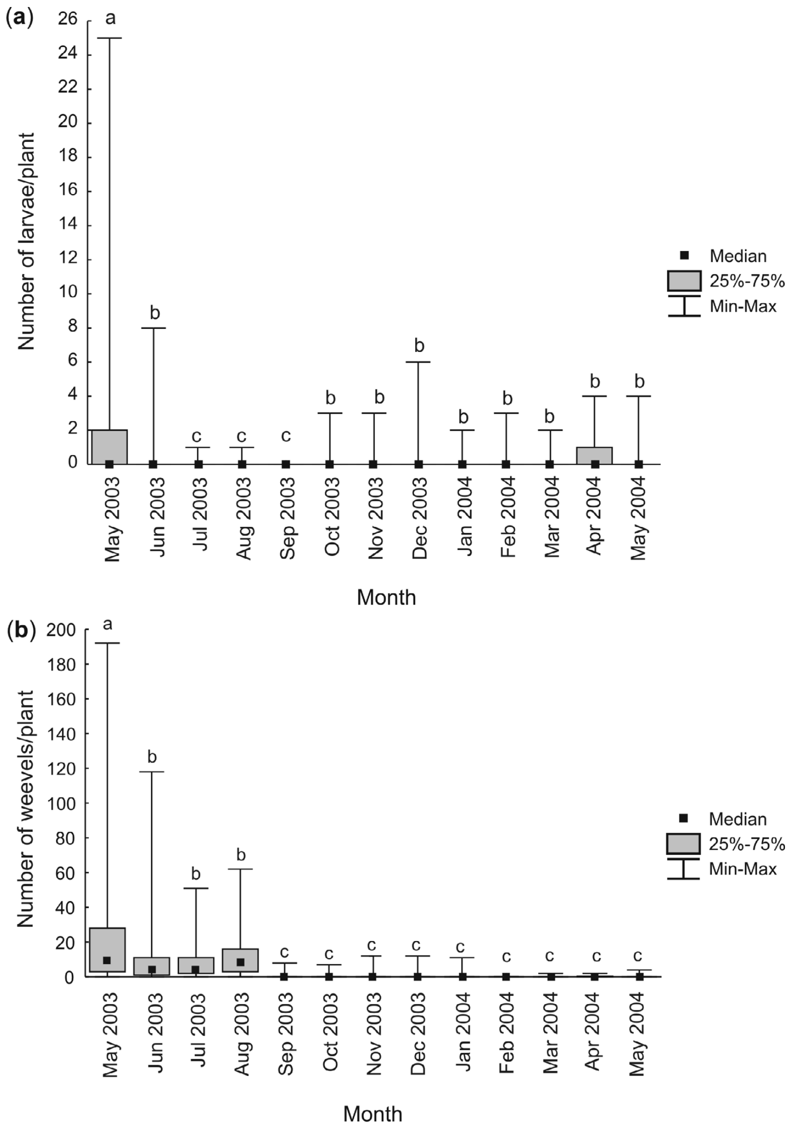

Fig. 5. Median number of larval (**a**) and adult (**b**) *Neohydronomus affinis* weevils per *Pistia stratiotes* plant sampled at Cape Recife over a 12-month period (n = 100 plants). Medians not followed by the same letter denote significant differences ($P < 0.0001$).

been visited at least once per year between 2004 and 2011 as part of an annual waterweed survey by Rhodes University. During this time, the percentage cover of *P. stratiotes* on the Cape Recife settlement ponds has not exceeded 5 % and more than 80 % of the plants showed weevil damage (Hill, unpublished data). The situation at Cape Recife contrasts that at Sunset Dam in the Kruger National Park and a reservoir in Mmabatho in North West Province where initial spectacular clearing of water lettuce by the weevils was followed by yearly resurgences of the weed that covered the entire surface of these water bodies (Cilliers 1991). At these, and a number of other sites in South Africa, the weevil required five to seven years to deplete the water lettuce seed bank to the extent that the weed never covered more than 5 % (Cilliers *et al.* 2003). Datta & Biswas (1970) showed that conditions of low oxygen and/or high carbon dioxide in the substrate, such as that found in sewerage settlement ponds, prevented the germination of *P. stratiotes* seeds. Although not measured, this is likely to be the case at the Cape Recife site, as it was in Benin, where *P. stratiotes* was successfully controlled within two years due to a lack of germination (Ajuonu & Neuenschwander 2003).

The establishment of a biological control agent usually relies on the release of large (thousands), healthy populations of the agents onto healthy plants in the field (Memmott *et al.* 1998; Hill & Olckers 2001). In this study, a small number of the agent (240 individuals) was released at the worst time of year (winter) onto healthy plants growing under highly eutrophic conditions. Despite this, the agents were able to establish and effect complete control at this site, where the weed no longer poses a problem and no other control methods are required to maintain weed populations.

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