

Distribution and impact of the Asian seed beetle, *Megabruchidius* tonkineus (Pic, 1904) (Coleoptera: Chrysomelidae: Bruchinae) on *Gleditsia triacanthos* L. seeds in South Africa

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Honey locust, *Gleditsia triacanthos* (Fabaceae) is a fast-growing deciduous tree native to North America. The tree has been introduced worldwide and has become invasive in South Africa. In 2017, it was listed as one of the nine fastest-spreading weeds in South Africa. Once established, it competes with and displaces indigenous species and dense stands can significantly reduce stream flow. The Asian seed-feeding bruchid, *Megabruchidius tonkineus* (Coleoptera: Chrysomelidae: Bruchinae) was introduced into South Africa and has subsequently taken honey locust as a host plant. The beetle damages honey locust seeds and is therefore considered a biological control agent, despite no host-specificity or impact studies having been conducted. This study shows that *M. tonkineus* has established across the entire *G. triacanthos* population range in South Africa, damaging approximately 9% of seeds. Laboratory studies show that *M. tonkineus* completes its larval development in the seeds of *G. triacanthos* in about 66.80 ± 0.69 SE days before emerging. This study is the first to describe the distribution and impact of this alien insect in South Africa.

Gleditsia triacanthos L. (Fabaceae) (honey locust) is a fast-growing and deciduous tree native to the United States of America, which has been introduced around the world as an ornamental plant (Isely 1975; CABI 2021). In South Africa it has become invasive and is regarded as one of the nine fastest-spreading weeds; it is a significant environmental threat to indigenous species by creating dense monocultures along watercourses and roadsides (Henderson and Wilson 2017; CABI 2021; Figure 1). The tree is listed under the 1b category of the National Environmental Management: Biodiversity Act 963 (NEMBA, Act 10 of 2004) Alien and Invasive Species Regulations (Department of Environmental Affairs 2014). Category 1b alien invasive species are to be removed on site but do not have national management programmes dedicated to their eradication. Since chemical or mechanical methods are currently not used to manage *G. triacanthos*, biological control has the potential to offer a sustainable cost-effective management option.

In South Africa, the Asian seed-feeding bruchid, *Megabruchidius tonkineus* (Pic, 1904) (Coleoptera: Chrysomelidae) (Figure 2) has been recorded eating the seeds of *G. triacanthos* (Martin 2021) and is considered as a biological control agent (Klein 2011; Zachariades 2021). The exact date and location of its unintentional introduction is unknown, although it has been suggested that the most likely date of introduction was the early 2000s (Di Iorio 2015; Zachariades 2021). *Megabruchidius tonkineus* was not officially released as a biological control agent despite being considered, rather it was found established in the field after research in quarantine had already been initiated on it; however, the distribution, hosts, and impacts of *M. tonkineus* were never determined.

Megabruchidius tonkineus is native to the northern region of Vietnam where it is associated with Gleditsia australis Hemsl. ex. FB Forbes, Hemsl. (György and Germann 2012; Yus-Ramos

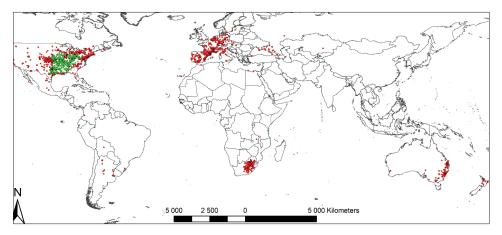


Figure 1. Global distribution of *Gleditsia triacanthos* (Green dots = native range; Red dots = invaded range). Adapted from the Global Biodiversity Information Facility (GBIF 2021), South African Plant Invader Atlas (SAPIA) (Henderson 2007) and Living Australia database (Atlas of Living Australia).

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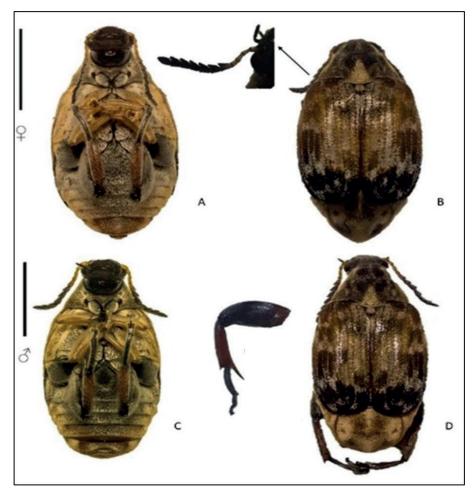


Figure 2. Megabruchidius tonkineus (Coleoptera: Chrysomelidae: Bruchinae) adults, Female (A and B) plus a magnified image of the diagnostic antenna is shown between the images; male (C and D) and a magnified image, clearly showing the diagnostic mucro on the apex of the metatibia. Scale bars: 1 mm

2014), a tree native to southern China and Vietnam. The beetle has also been reported from Europe (Bulgaria, Croatia, France, Germany, Greece, Hungary, Romania, Russia, Serbia and Switzerland), South America (Argentina and Chile), Australia, and Africa (South Africa) (Yus-Ramos et al. 2011; György and Germann 2012; Gavrilović and Savić 2013; Korotyaev 2016; Fernández et al. 2017; Kurtek et al. 2017; Pintilioaie et al. 2018; Eow et al. 2023). All the available literature suggests the larvae of *M. tonkineus* are only able to complete their reproductive cycle on species that are within the genus *Gleditsia* (Eow et al. 2023). Adults, which are pollen feeders, have however been collected from a variety of species including the flowers of marigolds (*Tagetes* spp.) (Asteraceae) (Korotyaev 2011).

In the field, females lay eggs singly on the mature pods of *G. triacanthos* both while the pods are still on the tree and also on pods that have fallen to the ground. Females preferentially oviposit where pods have been damaged, such as existing exit holes, or broken edges (pers. obs). If given the option, females will also oviposit directly onto seeds. At room temperature $(23 \pm 1.5 \text{ °C})$, eggs hatch after approximately 14 days. The exploratory larvae do not bore into the seeds immediately after hatching and may move around within the seed pod (György 2007; Salgado Astudillo 2021). Once a larva enters a seed, it will mine nearly all the available reproductive seed material. Occasionally two larvae may be recorded in a single seed (pers. obs.). The development from egg to adult takes approximately 68 days (Salgado Astudillo 2021).

This manuscript reports on the abundance, distribution, and impact of *M. tonkineus* in South Africa.

Surveys for *M. tonkineus* were conducted at 231 sites across South Africa; however, at only 60 could sufficient pods be collected for analysis. Surveys took place along roadsides within the plant's known distribution which included the Eastern Cape, Northern Cape, Free State and KwaZulu-Natal provinces (SAPIA 2021). The tree is abundant and easily distinguishable from other invasive trees in the grassland biome, making populations easy to locate. Surveys were conducted during summer and winter when mature seed pods were still on the trees and therefore the highest likelihood of finding insects within the pods.

Twenty mature seed pods were collected from each tree of which ten were picked from the tree and ten which had fallen to the ground. The two groups were selected to determine if there were differences in insect numbers between hanging and fallen pods. As no difference in insect numbers was found between the two groups, the two sets of pods were amalgamated. In addition, four soil cores using an auger (diameter 7.5 cm × 20 cm deep) were taken per site and the soil samples were passed through a sieve (5 mm), to remove fine particles, leaving only seeds and large particles in the sieve. Gleditsia triacanthos seeds could be clearly distinguished as they were quite large ($\sim 1 \times 0.75$ cm). This was firstly done to determine the number of seeds in the soil seedbank but also to determine if *M. tonkineus* was able to survive in seeds within the soil. All pods, and seeds collected from the soil bank, were stored inside paper bags and returned to the laboratory. In the laboratory, the pods and the seeds collected from the soil were transferred into individual plastic cages (20 cm \times 20 cm \times 17 cm) to record the presence and abundance of *M. tonkineus*. The pods were kept in the cages for two months until all the adults had emerged and before a second generation could start breeding. The emerged adults were removed from the plastic cages and the remaining seeds were processed to examine the number of damaged, deformed, and healthy seeds. The emerged adults were counted and identified using György (2007). A selection of the emerging adults was sent to the National Collection of Insects – Agricultural Research Council (ARC-PHP) Biosystematics for full identification, to confirm that the species was *M. tonkineus*. The remaining voucher specimens are housed at the Centre for Biological Control (CBC), Rhodes University and referred to by Rhodes University (RH) accession numbers e.g., Rh1300.

Megabruchidius tonkineus was present at 47 of the 60 sampled sites. The mean percentage of seeds that was damaged by *M. tonkineus*, from all the sites sampled in South Africa, was 9.2 \pm 1.3 SE. Of the remaining seeds, 10.7 \pm 1.70% were not fully developed and regarded as unviable, while 80.1 \pm 1.3% of seeds were regarded as healthy (seeds where the beetle had not caused any damage and would probably grow under favourable conditions) (Figure 3). No adult emerged from the seeds collected in the soil.

The results of the survey indicate that *M. tonkineus* is already widely distributed in South Africa. This is not surprising, as the species is known to spread quickly (Di Iorio 2015). Based on its current distribution, the insect can probably survive under a variety of environmental conditions, including the harsh highveld winters, during which temperatures regularly fall below 0 °C and over 100 frost days occur, as well as the warmer subtropical climates of KwaZulu-Natal province (Schulze 1997; Figure 3).

The biology of *M. tonkineus* in South Africa and potential damage to any other plant species still need to be determined. As *M. tonkineus* originates from Asia while *G. triacanthos* is from North America, this does not follow the principles of classical biological control and makes the relationship a new association. While Chrysomelidae are often regarded as excellent biological control agents, within Bruchinae they have a comparatively low

percentage of biological control success (Syrett et al. 1996). In South Africa, nine Bruchinae species have been considered as biological control agents; four were never released (three rejected, one shelved), one failed to establish, three have only contributed trivial control to their respective targets and only one has had considerable impact. Algarobius prosopis (Le Conte) (Coleoptera: Chrysomelidae: Bruchinae) was released in 1989 to reduce viable seed numbers of Prosopis. The agent spread throughout the distribution of the target weed and despite being significantly impacted by livestock, which eat the pods, its level of control is, in general, significant (Kleinjan et al. 2021; Zachariades 2021). In the absence of livestock, seed damage levels in pods on the ground often exceed 90% (Kleinjan et al. 2021). However, this is not always sufficient to bring about management; for example, Hoffmann and Moran (1991) showed that the 98% seed reduction in Sesbania punicea (Cav.) Benth. (Fabaceae) caused by the weevil Trichapion lativentre (Beguin-Billecocq) did not lead to a decline in the density of mature plants.

The current low levels (9%) of damage caused by *M. tonkineus* on *G. triacanthos* in South Africa suggest that the plant is likely to keep increasing in density and distribution in the absence of further management. Therefore, further biological control options should be considered, including agents which target the plant's reproductive structures.

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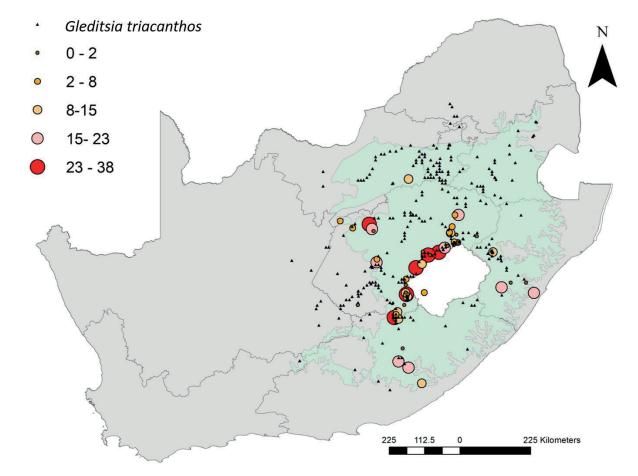


Figure 3. Relative abundance of *Megabruchidius tonkineus* (Pic, 1904) (Coleoptera: Chrysomelidae: Bruchinae) at recorded sites in South Africa, where the black triangles represent the distribution of *Gleditsia triacanthos* L (Fabaceae), according to the SAPIA database (Henderson 2007) and the coloured circles represent the percentage of damaged seeds caused by *M. tonkineus* from the 60 sampled sites. Light green colour indicates Grassland biome where the tree is most problematic (adapted from Mucina and Rutherford 2006)

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