# Seedling recruitment, seed persistence and aspects of dispersal ecology of the invasive moth vine, *Araujia sericifera* (Asclepiadaceae)

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Abstract. We investigated germination, seedling emergence and seed persistence of the invasive moth vine (Araujia sericifera Brot.) under controlled and field conditions in south-eastern Queensland. Fresh seed showed high viability (99.5%) and germinated readily, with 97.2% of all recorded germinations occurring within 14 days. Mean germination rates ranged from 74 to 100% across the range of temperature (10/20°C, 15/25°C and 20/30°C alternating 12-h thermoperiods) and light (0- and 12-h photoperiods) treatments. Germination was significantly reduced only under cool, dark conditions. In a 24-month field experiment, seedling emergence was greatest for seeds buried at 1 cm (mean = 96.3%), intermediate for seeds buried at 5 cm (mean = 62.7%) and least for surface-sown seeds (mean = 30.7%). Seed persistence under field conditions was low, declining rapidly to 3.9% at 6 months and to 0.67% at 24 months. Moth vine's capacity to germinate readily and the rapid depletion of seeds under field conditions indicate that the species has a transient seed bank in south-eastern Queensland. In an additional experiment testing the capacity of seeds for secondary dispersal by water, the mean floating time of seeds was 15.4 days, suggesting that water could act as a secondary dispersal vector, contributing to long-distance dispersal. We recommend that surveillance methods for detecting moth-vine populations should consider both water-flow patterns and wind direction.

## Introduction

Vine species are a significant, but generally poorly understood, group of environmental weeds. In south-eastern Queensland they comprise 13 of the top 50 invasive species (Batianoff and Butler 2002), and include species such as *Macfadyena unguis-cati*, *Asparagus* spp., *Aristolochia elegans*, *Anredera cordifolia*, *Cardiospermum grandiflorum*, as well as *Araujia sericifera*, the focus of this study. For most of these species there is insufficient ecological information to enable the development and implementation of effective management strategies.

Moth vine (*Araujia sericifera* Brot.) (Syn. *A. hortorum* E.Fourn, *Pysianthus albens* Mart., *A. sericofera* Brot.) (Asclepiadaceae) is native to southern Brazil and Argentina (Forster and Bruyns 1992). It was originally introduced to gardens for its attractive white flowers and its curiosity value as a moth trap (Stearns 1887). It is listed as invasive in Australia (Kleinschmidt and Johnson 1979; Muyt 2001), New Zealand (Ward *et al.* 1999; Coulston 2002), Spain (Carretero 1989) and North America (Bellue 1948; Spellman and Gunn 1976). Moth vine is also recorded as naturalised in other countries (Forster and Bruyns 1992), including southern Africa (Dyer 1975), Israel (Danin 2000) and Turkey (Altinözlü and Dönmez 2003).

In New Zealand the species poses a particularly serious weed problem in the North Island, where it is present in all regional areas, and has been given the highest weed-potential ranking in both Northland and Auckland regions (Winks and Fowler 2000). In Australia, moth vine is invasive in coastal New South Wales and south-eastern Queensland, where it is ranked 26th in importance as an environmental weed (Batianoff and Butler 2002) and has naturalised throughout most of the region (Stanley and Ross 1986), forming persistent populations. It has limited distributions in Victoria where it is an emerging weed (Muyt 2001), and is found occasionally in South Australia (Pearce 1986) and the ACT (Muyt 2001). Habitats where it is most common include disturbed rainforest edges and remnants (Joseph 1999) and forestry plantations on red loamy soils (Everist 1981). Moth vine is also problematic in riparian areas, moist forests and woodlands (Muyt 2001), disturbed coastal lowlands (G. Vivian-Smith, pers. obs.), roadsides and wastelands (Everist 1981).

In severe infestations moth vine can form a dense canopy over existing vegetation, eventually dominating and sometimes crushing native vegetation and preventing native plant regeneration (Muyt 2001; Coulston 2002). Seedlings are reported to be shade-tolerant, growing rapidly

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once a canopy gap is formed (Coulston 2002). Moth vine is a twining, perennial vine with thin woody stems that grows to less than 12 m. Flowers are produced during summer and autumn. They mature to produce pear-shaped, 6–10 cm long, grevish-green fruit (a follicle), with coarse longitudinal ridges that split open to release several hundred seeds (n = 10 fruits, containing 300–550 seeds, mean = 421 seeds; G. Vivian-Smith, unpubl. data). Individual seeds weigh  $9.62 \pm 0.12 \,\mathrm{mg}$  (mean  $\pm \,\mathrm{s.e.}$ , n = 80; G. Vivian-Smith, unpubl. data). In south-eastern Queensland, mature (dehiscing) fruits have been noted on vines from December through to August, suggesting that seeds can be dispersed during much of the year. A silky coma is attached to each seed and acts like a parachute to aid wind dispersal. Anecdotal reports suggest that rapidly growing seedlings can take as little as 12 months to flower and set seed (Bellue 1948); however, 24-36 months has been reported in Australian weed populations (Muyt 2001).

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Wind is the predominant dispersal vector, but the species is also reportedly dispersed by water (Muyt 2001), probably because it is found colonising riparian and low-lying areas that may be infrequently inundated. Seeds of other species within the Asclepiadaceae have shown a capacity to remain both buoyant and viable for lengthy periods (weeks to months) (Edwards *et al.* 1994). A better understanding of the capacity of this species to be dispersed by water may help refine detection methods and lead to better weed management.

There are no published data on germination, seed-bank dynamics or dispersal of the species. Winks and Fowler (2000) reported seed persistence for up to 5 years, but cited no supporting data. A report from observations of field eradication efforts also suggests seeds may remain viable for up to 5 years (Coulston 2002); however, the authors indicated that the species was difficult to detect during follow-up monitoring, so it is possible that inputs to the seed bank from undetected plants continued during the eradication attempt. Seed-bank dynamics of other members of the Asclepiadaceae, including both milkweed, Asclepias syriaca L. (Oegema and Fletcher 1971), and rubber vine, Cryptostegia grandiflora Roxb. Ex R.Br (Grice 1996; Bebawi et al. 2003), suggest that a wide range of seed-dormancy and persistence patterns exists within the Asclepiadaceae. This variability provides little predictive power with regard to information that is critical for the development of effective management strategies for moth vine.

The primary aims in this study were to better understand recruitment of moth vine by (1) determining the germination response to temperature and light, (2) assessing levels of emergence from the seed bank in relation to different sowing treatments and (3) determining seed persistence in relation to burial treatments under field conditions. A secondary aim was to estimate the capacity of moth vine to be dispersed by water by determining the length of

time that seeds and mature fruits remained buoyant under controlled conditions.

#### Methods

Mature fruits were collected from a minimum of five individuals at each of two coastal lowland populations at Bulwer Island (27°24′20″S, 153°08′09″E) and Boondall (27°20′09″S, 153°04′39″E) in Brisbane during March 2002. Bulwer Island is a disturbed lowland dominated by broad-leafed pepper (Schinus terebinthifolius), lantana (Lantana camara) and tall exotic grasses. Boondall was also a lowland site, but dominated by Casuarina glauca, with infrequent Eucalyptus tereticornis, Passiflora suberosa and L. camara. For all experiments, with the exception of the buoyancy experiment, the very weakly attached coma fell off or was removed during preparation, to facilitate handling and experimental set up (a comparison of germination rates between seeds + coma and seeds without coma indicated that the presence of the coma did not influence germination) (Vivian-Smith, unpubl. data). A composite sample of seeds collected at Bulwer Island and Boondall was used in each experiment. Only filled seeds were utilised in each experiment.

#### Germination response

A factorial experiment (n = 5 replicates) testing the germination response to light (12-h photoperiod) and continuous dark conditions over a range of temperature treatments (12-h alternating thermoperiods of 10/20°C, 15/25°C and 25/30°C) commenced on 8 April 2002, approximately 2 weeks after fruits were collected. These thermoperiods reflect a range of diurnal temperature conditions found in south-eastern Queensland during most seasons. In each Petri dish, 20 seeds were placed on a filter paper (Advantec 2) on an inverted watch glass, with 8 mL of tap water added. The inverted watch glass enabled a reservoir of water to be maintained at the base of each Petri dish. Dark treatments involved wrapping Petri dishes in two layers of aluminium foil. All Petri dishes were randomly placed in clear, sealed plastic bags to create similar conditions of humidity for both light and dark (wrapped) Petri dishes. Additional water was added to each Petri dish on a weekly basis as required. Germination was recorded weekly until it ceased. Ungerminated seeds from the low-temperature treatment were removed and placed on clean filter paper in new Petri dishes with 8 mL of water in a cabinet at 15/25°C to determine whether dormancy was enforced by low temperatures.

#### Seedling emergence

Seedling emergence in response to sowing at 0-, 1- and 5-cm depths (n=10 replicates per treatment) was monitored in a field experiment at the Alan Fletcher Research Station (AFRS). Thirty seeds were sown per pot on 16 April 2002, using a standard commercial topsoil mix (Centenary Landscaping Pty Ltd) and methods adapted from Panetta (2000). There was no supplementary watering, and natural rainfall was the only source of water received. Pots (12 cm diameter) were sunk into the soil to 11 cm. A wooden lattice (4-cm strips, with  $4.5 \times 4.5 \text{ cm}$  spacing between the strips) was placed 18 cm above the soil surface and covered with 50% shadecloth. Field conditions aimed to simulate those likely to be experienced by seeds landing below a suitable host tree or low canopy. Plots were located within a rodent-proof fence to reduce the likelihood of seed predation by rodents. Emerging seedlings were recorded and then removed from pots at monthly intervals for 24 months.

## Seed persistence

Seed persistence was determined for seeds exposed to the same sowing treatments (0-, 1- and 5-cm depths) and field conditions as in the emergence experiment. Squares of nylon-mesh insect screen were buried 1 cm below the seeds to reduce the amount of soil from which

the propagules were later extracted. Persistence was determined by retrieving subsamples of five replicates (n = 30 seeds per replicate) of each sowing treatment every 6 months for 2 years and testing the seeds for viability. Viable seeds included seeds that were germinable as well as those that were assessed as dormant. Retrieval of seeds was assisted by passing the soil containing seeds through a 1-mm sieve. Decayed seeds (those that disintegrated when gently rubbed between thumb and forefinger) were counted and discarded. The remaining seeds were then placed in Petri dishes and subjected to germination tests, using the same methods as described for the germination experiment (15/25°C and a 12-h photoperiod). When no further germination was recorded, the remaining seeds were dissected longitudinally and embryos were visually assessed for viability with a dissecting microscope. Seeds decayed, or containing a decayed or discoloured (brown-yellow) embryo were considered non-viable. Seeds containing an embryo that was firm and white were considered viable.

#### Seed buoyancy experiment

A buoyancy experiment was conducted to determine flotation times for both seeds and fruits collected from Bulwer Island and Boondall populations. Experiments were undertaken in an experimental pond at AFRS. Experimental ponds are recommended as an improvement over traditional laboratory-based experimental systems for estimating buoyancy (Edwards et al. 1994); however, they are representative of a non-turbulent water body, rather than the full range of water flows that seeds are likely to experience in a natural system (e.g. in a flooded creek). Therefore, the floating times obtained in such an experimental system should be interpreted as maximal or upper-limit values likely to be achieved in a natural system. Propagules were placed in floating cages constructed from plastic containers covered with fine nylon mesh and attached to polystyrene floats, using methodology adapted from Edwards et al. (1994). Ten replicate cages each containing 30 seeds and 16 replicate cages containing individual fruits were used. Assessments of the numbers of propagules floating were made for each replicate daily during the first 5 days of each experiment and every 3-5 days thereafter.

## Data analysis

Germination, emergence and seed persistence data were strongly skewed and no transformations were able to improve their normality to meet assumptions of ANOVA. As a result, the non-parametric Kruskal–Wallis test was used to determine treatment effects for these data. For the seed persistence experiment we used the non-parametric Friedman's

test to detect differences in seed persistence across the 6-, 12-, 18-, and 24-month sampling intervals within each sowing (burial depth) treatment. Data for seed and fruit buoyancy experiments were analysed by using survival analysis (Kaplan–Meier estimates in conjunction with a stratified Cox proportional hazard model) to determine mean floating (survival) times and to determine whether fruits and seeds showed different survival functions. All data analysis was conducted with Systat<sup>®</sup> 9.

#### Results

## Germination

Moth-vine seeds germinated rapidly, with a mean germination level of 70% recorded within 7 days. Germination virtually ceased after 14 days, with only 2.6% occurring beyond this time. Germination rates showed a significant (Kruskal–Wallis:  $H=20.449,\ P=0.001$ ) response to temperature and light treatment combinations. However, this was primarily due to the considerably lower germination level (74%) in the cool, dark treatment (Fig. 1). In all other treatments germination levels were consistently high and ranged from 97 to 100% (Fig. 1). Of the 32 (16%) remaining ungerminated seeds from the cool, dark treatment, 31 germinated within 7 days when placed in the intermediate  $15/25^{\circ}$ C, 12-h photoperiod treatment.

## Seedling emergence

Seedling emergence commenced within 3 weeks of sowing under field conditions, reaching a maximum rate between 21 and 60 days of sowing. After this period few seedlings were detected (Fig. 2), with only 2.1% emergence occurring during the remaining 22 months of the experiment. Seedling emergence responses were significantly (Kruskal–Wallis: H = 20.364, P < 0.001) different for each sowing treatment at both 6 and 24 months. At 24 months, emergence was greatest for seeds buried at 1 cm (mean = 96.3%), intermediate for seeds buried at

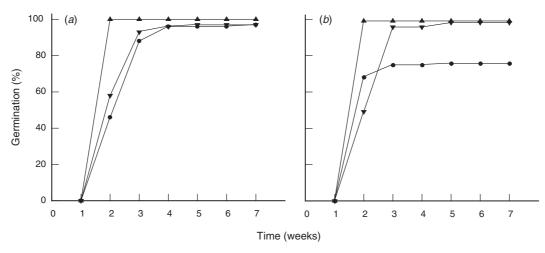
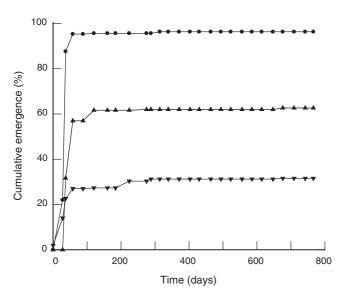


Fig. 1. Cumulative germination response of *Araujia sericifera* temperature.  $\bullet$ , 10°C/20°C;  $\blacktriangle$ , 15°C/25°C;  $\blacktriangledown$ , 20°C/30°C alternating 12-h thermoperiods under (a) 12-h photoperiod and (b) continuous dark conditions.

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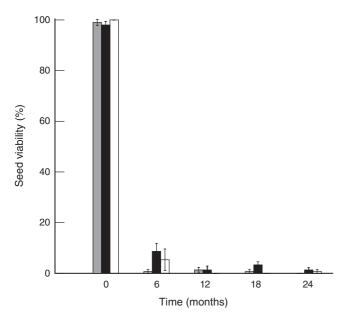
**Fig. 2** Cumulative emergence of *Araujia sericifera* seedlings under field conditions over 766 days. ●, 1-cm burial; ▲, 5-cm burial; ▼, surface-sown seeds.

5 cm (mean = 62.7%) and least for surface-sown seeds (mean = 30.7%) (Fig. 2).

## Seed persistence

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The number of viable (dormant + germinable) seeds present at each sampling period declined from an initial value of 99% to values less than 10% (mean values: buried 1 cm = 0.67%; buried 5 cm = 8.67%, surface = 5.33%) at 6 months (Fig. 3).



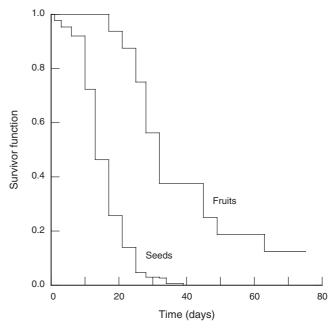
**Fig. 3.** Percentage of viable *Araujia sericifera* seeds remaining at 0-, 6-, 12-, 18- and 24-months for three sowing depths: 1 cm (gray bars), 5 cm (black bars), 0 cm (open bars). Error bars represent s.e.m.

For buried seed, this was primarily due to the large numbers of seedlings emerging during this period (see Fig. 2). At 12 months, on average only 0.89% of seeds remained. At 24 months, when the experiment was terminated, only 0.67% viable seed remained (mean values: buried 1 cm = 0.0%; buried 5 cm = 1.33%; surface = 0.67%) (Fig. 3). Seeds retrieved showed low levels of both germinability and dormancy, suggesting that enforced and/or induced dormancy may occur, but at low frequencies for this species.

Comparisons of seed viability among sowing treatments showed no significant (Kruskal–Wallis: H=7.43, P=0.024) differences, with the exception of the 18-month sampling (mean values: buried 1 cm = 0.67%, buried 5 cm = 3.33%, surface = 0%). Within each sowing treatment, no significant differences were detected by Friedman's test when comparisons of viability were made among the 6-, 12-, 18- and 24-month sampling periods.

## Seed buoyancy

Moth-vine seeds had a mean floating time of 15.4 days, although there was significant (P = 0.036) variation in mean floating times among cages (range 12.3–18.4 days). This suggests a significant capacity for water dispersal, but with susceptibility to variation resulting from localised conditions. Although moth-vine fruits floated significantly ( $\chi^2 = 44.362$ , d.f. = 1, P = 0.016) longer than seeds (mean survival time = 38.7 days) (Fig. 4), our observations indicate that fruits generally remain firmly attached to the parent



**Fig. 4.** Survivor (where survivors = individuals remaining floating) probability curves for *Araujia sericifera* seeds and intact fruits expressed as unadjusted Kaplan–Meier stepped functions, showing different survival functions for seeds and intact fruits.

vine, where they split open and disperse seeds. The pods used in this experiment were ripe, but had not dehisced. Several split open during the course of the experiment, whereas others remained intact for the duration of the experiment and appeared to have viable seeds on termination (although this was not tested). Therefore, the long floating times recorded for the fruits of this species should not be interpreted as a measure of downstream dispersal capacity, although it is possible that this may occur during flooding events when branches/vines containing fruit may be transported on rafts of vegetation. Many (82%) moth-vine seeds germinated in the water, but few seedlings (10.3%) survived 30 days of immersion, suggesting that dispersal following temporary, rather than prolonged, flooding could lead to transport of seedlings/seeds to microsites suitable for establishment.

## Discussion

Results from our germination experiment indicated that moth-vine seeds have high viability, and do not have specific germination requirements, as demonstrated by the high germination rates that were observed under a wide range of temperature and light conditions. Although germination was somewhat reduced (74%) under cool, dark conditions, ungerminated seeds germinated quickly when placed in more favourable conditions, indicating that low temperatures can result in an imposition of enforced dormancy in a proportion of seeds. The germination responses of moth vine show strong similarities to those of the weed of national significance, *Cryptostegia grandiflora* (rubber vine) Asclepiadaceae, a species that also germinates rapidly to levels of approximately 90% under a range of temperatures (Grice 1996).

Moth-vine seedling-emergence patterns indicated that recruitment is favoured by shallow seed burial, but is still considerable (62.7%) from depths of 5 cm. This suggests that disturbance leading to incorporation of seeds within the soil profile could favour seedling recruitment. Our germination experiments indicated that no light requirement exists. Surface-sown seeds showed the lowest levels of seedling recruitment (30.7%). Although low, this could still be sufficient to generate a high density of seedlings within a natural population. Losses of surface-sown seeds to predation are unlikely to have been significant, since most of the remaining ungerminated seeds were recovered but recorded as decayed/empty seeds during the seed-retrieval process. It should also be noted that during the initial growing season (April–June 2002) of our experiment, rainfall was low (AFRS rainfall: 138.5 mm; 115-year average: 211.85 mm). Therefore, it is possible that with closer to average rainfall, greater levels of seedling recruitment from surface-sown seeds could occur.

Our results suggest that moth vine is characterised by a transient (<12 month) seed bank, one that is largely depleted through seedling emergence and retains less than 1% of viable

seeds at 12 months. Very low levels (0.67%) of dormant, but viable, seeds were detected at 24 months, suggesting that induced dormancy may occur, but at a very low frequency. Although it is possible in natural populations that induced dormancy could lead to reinfestation as a result of seedling recruitment from the *in situ* seed bank after 24 months, we believe that the probability of this occurring would be quite low. Further work to determine the densities of dormant and germinable seeds present in *in situ* seed banks is required to validate this.

The 5-year estimates of seed persistence from New Zealand workers (Winks and Fowler 2000; Coulston 2002) are considerably greater than the values suggested by our experiment. We are unclear as to how these estimates were derived; it is possible that the New Zealand field situations have included seed inputs from missed adults during the eradication process. However, in the case of extensive/dense infestations producing large inputs to the seed bank, a low frequency of induced dormancy might contribute to a more persistent seed bank.

From a management perspective, the transient (<12 month) nature of the moth-vine seed bank indicates a potential weakness in the species' life cycle that could be exploited during management operations. In this respect, moth-vine seed bank dynamics show strong similarities to those of rubber vine (Grice 1996; Bebawi et al. 2003) and other invasive vines that are currently under investigation (e.g. Vivian-Smith and Panetta 2004). Our study suggests that maintenance of a seed bank is dependent on frequent (i.e. yearly) additions. The highly germinable nature of moth-vine seeds, in conjunction with high growth rates, can result in plants that are capable of contributing to the seed bank in 12–18 months. This suggests that short-term (24 month) control efforts that kill mature plants prior to seed production, in addition to seedlings emerging from the seed bank, could lead to successful control at a local level, providing inputs to the seed bank from external sources are also prevented.

In locating external seed sources that are likely to contribute to reinfestation, consideration should be given to patterns of water flow and prevailing wind directions during the seed-production period. Results from our buoyancy experiment suggest that for flood plain or riparian infestations it is advisable to locate and target upstream populations, as water is probably a secondary seed dispersal vector. Although wind dispersal was not investigated in this study, dispersal studies of related species may provide some useful information. For example, Cappuccino et al. (2002) demonstrated that seeds of the North American invasive vine, Vincetoxicum rossicum (Asclepiadaceae), a species of similar stature with comose seeds slightly smaller than those of Araujia sericifera, travelled up to 18 m from the release point when winds averaged 11.2 km h<sup>-1</sup>. Such information could be utilised when developing searching models or strategies for moth vine. Given moth vine's capacity for longdistance dispersal via both wind and water, a continued risk of reinfestation exists and longer-term surveillance to detect other infestations would be advisable.

Further research to enable development of effective strategies for managing moth vine is required. Areas that would benefit from future study include (1) the demography of moth-vine populations under natural conditions, including the size of *in situ* seed banks, and (2) integrated management that incorporates fire, in addition to the mechanical and chemical methods currently employed (e.g. Armstrong 1998; Muyt 2001; Coulston 2002). Additional work on seed persistence of New Zealand populations of moth vine would also be valuable.

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