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A novel stem-applied herbicide-capsule methodology for control of the invasive cactus *Cereus uruguayanus*

Leisa Bradburn[A,B,*], Shane Campbell[B], Vincent Mellor[B] and Victor Galea[B]

For full list of author affiliations and declarations see end of paper

***Correspondence to:**
Leisa Bradburn
Queensland Department of Agriculture and Fisheries, Gatton, Qld, Australia
Email: leisa.bradburn@daf.qld.gov.au

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ABSTRACT

Cereus uruguayanus R.Kiesling is a naturalised, non-opuntioid cactus that has formed large and dense infestations at several locations in the northern Australian rangelands. Of the Cactoideae subfamily, it typically grows into a large, spiny, single- or multi-stemmed candelabra-shaped cactus. While not classified as a Weed of National Significance (WONS) in Australia, it is causing increasing concern, with some local governments declaring it under their local laws. Recent research has identified several herbicides that control *C. uruguayanus* by using a range of techniques, including basal-bark, cut-stump, foliar, and stem injection applications. Of these, stem injection is one of the recommended options where weeds grow among desirable vegetation because the risk of non-target damage is minimised. This study evaluated *C. uruguayanus* control using a range of encapsulated dry-formulation herbicides from BioHerbicides Australia (BHA Pty Ltd), using their novel stem-implantation system (Injecta®). In May 2018, an experiment was established in central Queensland to compare the efficacy of six encapsulated herbicides (aminopyralid + metsulfuron-methyl, clopyralid, glyphosate, hexazinone, metsulfuron-methyl, triclopyr + picloram) against an untreated control. Glyphosate was the fastest-acting herbicide, followed by aminopyralid + metsulfuron-methyl and metsulfuron-methyl, with triclopyr + picloram much slower to act. Nevertheless, all four herbicides eventually caused high mortality (≥85%). In contrast, clopyralid and hexazinone were ineffective at the applied rates. Future research is recommended to compare the cost effectiveness of this system against other techniques, particularly those used for stem-injection applications.

Keywords: cactus, capsule, *Cereus uruguayanus*, control, granular, herbicide, implant, invasive, night blooming cereus, Peruvian apple cactus, weed, Willows cactus.

Introduction

Cereus uruguayanus R.Kiesling is a native cactus (Cactaceae family) from South America (Argentina, Brazil and Uruguay) that has become a naturalised invasive weed in Australia (Forster and Schmeider 2000; Campbell *et al.* 2021; Biosecurity Queensland 2022). Common names include apple cactus, candelabra cactus, hedge cactus, night-blooming cereus, Peruvian apple, torch cactus, and Willows cactus (Hosking *et al.* 2007; Central Highlands Regional Council 2015). Scientifically, it was referred to as *C. peruvianus* Mill., but was renamed *C. uruguayanus* by Kiesling (1982; Forster and Schmeider 2000).

Cereus uruguayanus is described in llifle Encyclopedia of Living Forms (2005) and Biosecurity Queensland (2022). A columnar candelabra-shaped cactus, it can grow up to 8 m, and produces blue-green to dull-green single or multiple vertical stems and branches generally containing six to nine spiny ribs (Fig. 1). Flowers are white and funnel-like, and open mostly at night, and the large red fruit contain hundreds of seeds. Monstrosity or fascination of the stem of some *C. uruguayanus* is observed occasionally in infestations in Queensland and occurs when the apical meristem divides in an abnormal manner, leading to a different appearance (Forster and Schmeider 2000).

In Australia (particularly Queensland), *C. uruguayanus* has been a popular ornamental plant, but often escapes into the surrounding environment (Forster and Schmeider 2000; Hosking *et al.* 2007). Its fleshy fruits are attractive to animals (particularly birds), and

Fig. 1. *Cereus uruguayanus:* (*a*) mature, multi-branched cacti plant, (*b*) flower in full bloom, and (*c*) fruit.

endozoochory has been suggested as a primary dispersal mechanism (Forster and Schmeider 2000).

Several large infestations of *C. uruguayanus* occur in central and southern inland Queensland, with smaller outbreaks elsewhere (Biosecurity Queensland 2022). Hosking *et al.* (2007) listed *C. uruguayanus* as naturalised in New South Wales in 2007 after it was positively identified at a few locations. *C. Uruguayanus* is not considered a Weed of National Significance (WONS), nor a declared weed in any state or territory, although it has been declared a priority pest species by some local governments (Central Highlands Regional Council 2015; Campbell *et al.* 2021; Biosecurity Queensland 2022).

Several herbicides are affective against *C. uruguayanus* using a range of application techniques, including basal-bark, cut-stump, foliar, and stem injection applications (Campbell *et al.* 2021; Biosecurity Queensland 2022). Of these, stem injection is recommended to minimise the risk of non-target damage where *C. uruguayanus* grows among desirable vegetation (Vitelli and Pitt 2006; McKenzie *et al.* 2010). A new technique has also been developed, whereby dry formulations of encapsulated herbicides are implanted into the stem (Goulter *et al.* 2018; Galea 2021). This has the advantages of being target specific, and greatly reducing operator risk by minimising contact with the herbicide. It has been tested successfully on a range of woody weeds, with other species being under investigation (Goulter *et al.* 2018; Limbongan *et al.* 2021; O'Brien *et al.* 2022).

This study aimed to test stem implantation to control *C. uruguayanus* by using a range of encapsulated dry-formulation herbicides from BioHerbicides Australia (BHA Pty Ltd), including glyphosate and hexazinone that have previously proven effective using traditional stem-injection approaches.

Materials and methods

Site details

The trial site (23°44′S, 147°32′E) was located on a cattle property near Willows, Queensland, Australia, within the

Brigalow Belt, which characteristically comprises clay soils, deep depressions (gilgais) and a dominance of *Acacia harpophylla* F. Muell. Ex Benth. This site had a medium-density *C. uruguayanus* infestation growing primarily among *A. harpophylla*, several *Eucalyptus* spp. and *Carissa ovata* R.Br. Plants were initially healthy as they had not been subjected to control activities (e.g. mechanical or chemical). During the 76-week study duration, the area was grazed by cattle and was accessible to the public for fossicking of semi-precious gems. However, these activities did not compromise trial integrity.

Experimental design

The experiment was conducted between 17 May 2018 and 2 November 2019 using a randomised complete block design (four replications), and six herbicide treatments plus a control (Table 1). Experimental units were parallel rows of 15 *C. uruguayanus* plants ranging from 20 to 35 m in length, depending on plant density. The first plant in each experimental unit was labelled, identifying the rep and treatment, all other treated plants were paint-marked for identification. Treated plants were at least 2 m apart, with a minimum 5 m buffer between treatments. To ensure consistency, selected plants were healthy, with no signs of physical damage, had a minimum stem circumference of 8 cm for treatment application, and were a minimum of 1 m and a maximum of 5 m high. Plants with multiple main stems and/or atypical branching or signs of monstrous and cristate stems were avoided in preference for those with single main trunks and typical branching (Fig. 1). On average, plants were 2.36 ± 0.04 (s.e.) m high, with a mean stem diameter of 18.59 ± 0.29 (s.e.) cm.

Treatment application

Selected herbicides (Table 1) were sourced from Bioherbicides Australia Pty Ltd, as dry formulations contained in size 0 Hypromellose capsules, and were a comprehensive representation of chemicals commonly used for control of woody weeds and some cactus species in Australia. An Injecta® applicator (BioHerbicides Australia Pty Ltd, Brisbane, Qld, Australia), consisting of a tube body, shaft and handle to which a cordless drill can be attached, was used to implant the capsules (Fig. 2*c*). The applicator was loaded with a magazine that can hold 30 capsules and wooden plugs (BioHerbicides Australia Pty Ltd, Brisbane, Qld, Australia) in tandem (Fig. 2*a, b*). Using this device, a hole was drilled into the plant stem via an 8 mm drill bit. The hole was automatically cleared of wood shavings, and through a smooth action of withdrawing and then pushing the drill forward, the capsule (Fig. 2*a*) and a plug were inserted together into the hole, sealing the capsule into the plant stem (Fig. 2*d*). The seal created by the plug allows plant sap to infiltrate the capsule and dissolve the herbicide. If a plug is not used to seal the implantation site, it is possible for the drill wound to oxidise and create scar tissue that can form a seal preventing herbicide uptake (Galea 2021).

The number of capsules inserted into each plant was based on stem basal circumference. A capsule was inserted for every 15 cm increment in plant circumference, as follows: 0–15 cm = 1 capsule, 16–30 cm = 2 capsules, 31–45 cm = 3 capsules, 46–60 cm = 4 capsules. When required, multiple capsules were implanted evenly around the stem. Capsules were inserted at the main stem base where there were generally fewer thorns, and where it was easier to avoid branches and access the stem.

To determine any adverse effect of the capsules themselves, 15 plants were implanted with a blank capsule containing perlite (Brunnings Australia, Dudley Park, South Australia), at the designated rates based on the stem circumference of individual plants.

Assessment

The trial was first assessed 4 weeks after treatment (WAT) and assessments continued every 4 weeks until 32 WAT.

Table 1. Herbicide treatments applied to *C. uruguayanus* using the Injecta® stem-implantation system.

Treatment number	Description	Dose (mg/capsule) of a.i. (active ingredient)	a.i. concentration (g/kg)	a.i. dose (mg/capsule)
T1	Control	Nil	Nil	Nil
T2	Di-Bak AM	Aminopyralid 155 & Metsulfuron-methyl 125	375	58.1
			300	37.5
T3	Di-Bak C	Clopyralid 450	750	337.5
T4	Di-Bak G	Glyphosate 350	700	245
T5	Di-Bak H	Hexazinone 350	750	262.5
T6	Di-Bak M	Metsulfuron-methyl 330	600	198
T7	Di-Bak TyP	Triclopyr 120 & Picloram 40	300	36
			100	4

Fig. 2. Injecta® herbicide capsule delivery method: (*a*) mechanised capsule applicator, magazine and wooded plugs, (*b*) capsule magazine being loaded with capsules and plugs, (*c*) herbicide capsules and plugs, (*d*) *C. uruguayanus* plant injected with capsule and plug.

A final assessment was conducted 76 WAT to confirm treatment effects. No monitoring of the trial occurred between the 32 and 76 WAT assessments.

At each assessment, percentage stem necrosis was recorded for all plants included in the trial, using a rating system of 0–10 with 0 = 0%, 1 = 1–10%, 2 = 11–20%, 3 = 21–30%, 4 = 31–40%, 5 = 41–50%, 6 = 51–60%, 7 = 61–70%, 8 = 71–80%, 9 = 81–90% and 10 = 91–100%. Plants that displayed no treatment effects were recorded as zero, whereas plants that developed necrosis were recorded with the corresponding percentage rating. At 32 and 76 WAT, all plants with a rating of 10 were also categorised as dead or alive based on the presence/absence of moisture and live growth following physical inspection of the stem. By 76 WAT, dead plants had either fallen over or were easily dislodged when assessed.

Data analysis

Data analyses were conducted using R (R Core Team 2022), by using Analysis of Variance (ANOVA) at $P < 0.05$. The assumptions of the general linear model were checked, and the data were transformed to better satisfy the assumptions. If significant treatment effects were present, estimated marginal means were calculated through the Estimated Marginal Means (emmeans) package (Lenth 2022), compared using Tukey's honest significant difference tests (HSD) and presented in a compact letter display table through the package Multcomp (Hothorn *et al.* 2008), to determine which treatments differed from each other at $P < 0.05$. This table groups treatments of similar performance and allows significant differences in performance to be quickly assessed.

For the necrosis data, observed results were first transformed by calculating the mean score of the 15 plants in each plot for each assessment date. The mean scores were then analysed by the method described above to identify any significant treatment differences.

For the mortality assessment, the proportion of dead plants was calculated for 32 and 76 WAT. These mortality proportions were then transformed using an arcsine transformation and then analysed using ANOVA. The estimated marginal means were back-transformed to the original scale after the *post hoc* testing had been completed.

Results

Climatic conditions

Mean minimum temperatures ranged between 4.5°C and 20.8°C in 2018 and between 7.1°C and 22.8°C in 2019. Mean maximum temperatures between 23.7°C and 37.1°C were recorded in 2018 and between 22.0°C and 37.2°C in 2019 (Bureau of Meteorology 2022; Fig. 3).

Rainfall during the trial period was minimal, with most months receiving below 30 mm (Fig. 4). During 2018, October was the only month when substantial rainfall was recorded. In 2019, March and April were the only months with substantial rainfall (Bureau of Meteorology 2022).

Treatment effects

Effect of blank capsules

Plants implanted with a blank capsule containing perlite remained healthy during the 76-week trial period.

Necrosis

Significant treatment differences ($P < 0.05$) were recorded in the level of necrosis observed on *C. uruguayanus* at all assessments (Fig. 5). However, irrespective of the treatment applied, initial signs of necrosis in the plant stem often varied in location, ranging from at the injection site, the top of the stem, in the middle of the stem or at the ridges of the stem (Fig. 6). In some plants, multiple points of necrosis occurred. For effective herbicide treatments, the necrosis progressively spread throughout the plant stem.

Untreated plants exhibited minimal necrosis, recording an average rating of 0.6 at 76 WAT. The initial plant response to herbicide treatments was limited, except for glyphosate (average rating of 4.1), which exhibited significantly higher ($P < 0.001$) necrosis than other treatments (average rating of ≤ 0.8) at 8 WAT. Necrosis remained consistently higher in the glyphosate treatment for most of the trial duration, and at the 76 WAT assessment had an average necrosis rating of 9.8 (Fig. 5).

Although necrosis presented less rapidly in the aminopyralid + metsulfuron-methyl and metsulfuron-

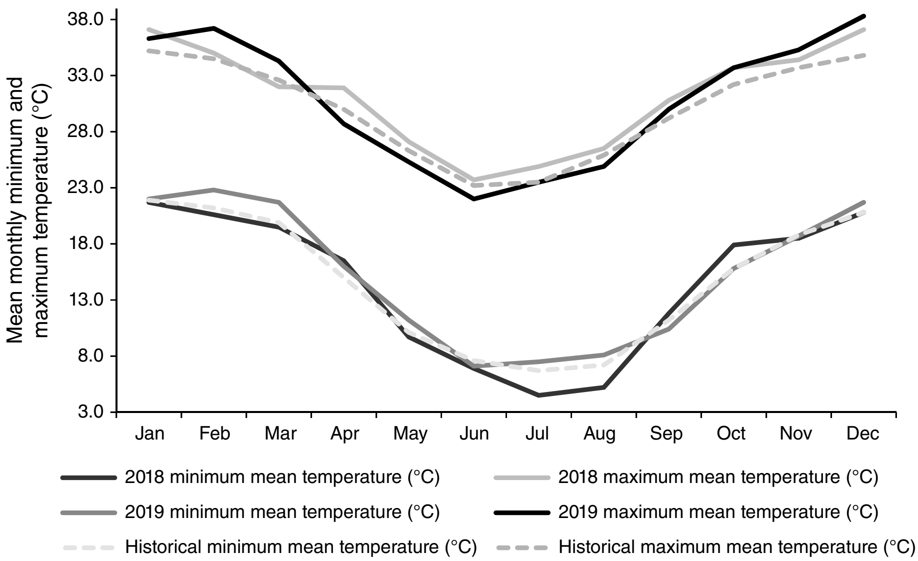

Fig. 3. Trial mean monthly minimum and maximum temperatures for 2018 and 2019 and historical climate statistics for Willows, Queensland, Station 035139 Lochington, Queensland (22.4 km away; Bureau of Meteorology 2022).

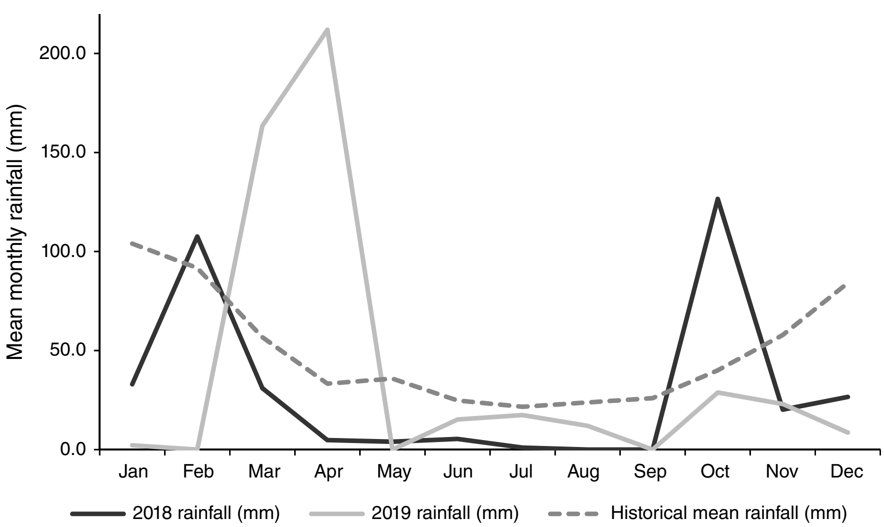

Fig. 4. Trial mean monthly rainfall for 2018 and 2019 and historical climate statistics for Willows, Queensland, Station 035215 Willow Gemfields, Queensland (0.2 km away; Bureau of Meteorology 2022).

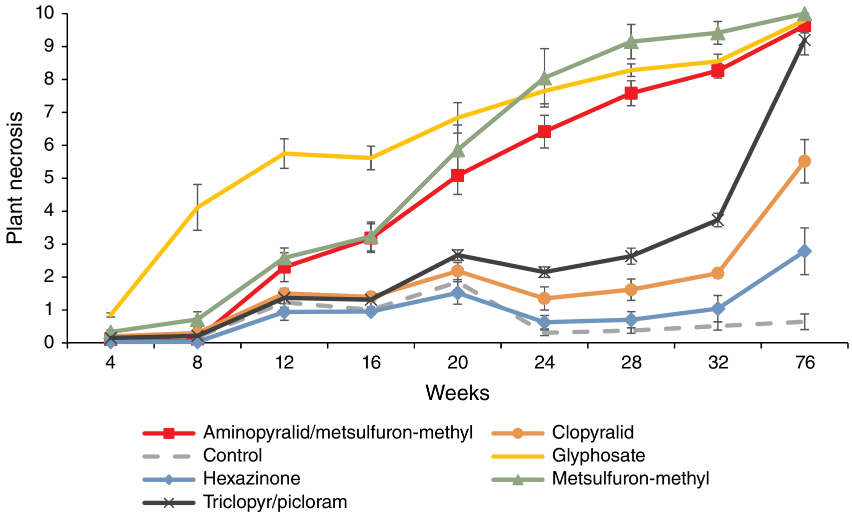

Fig. 5. *Cereus uruguayanus* plant necrosis rating score over the trial duration. Necrosis rating system: 0 = 0%, 1 = 1–10%, 2 = 11–20%, 3 = 21–30%, 4 = 31–40%, 5 = 41–50%, 6 = 51–60%, 7 = 61–70%, 8 = 71–80%, 9 = 81–90% and 10 = 91–100%. Error bars represent ± standard error of the means.

Fig. 6. *Cereus uruguayanus* necrosis: (*a*) necrosis at injection site, (*b*) necrosis at the top of the cactus, (*c*) necrosis at mid-stem, (*d*) necrosis at the ridges.

methyl treatments, it increased steadily from 8 WAT, and was not significantly different ($P > 0.05$) to glyphosate at 76 WAT, with average necrosis ratings of 9.6 and 10 respectively (Fig. 5). The triclopyr + picloram treatment was slower to act, with a necrosis rating of 3.7 at 32 WAT. However, the level of necrosis increased, and, by 76 WAT, it was not significantly different ($P > 0.05$) among the aminopyralid + metsulfuron-methyl, glyphosate and metsulfuron-methyl treatments, with an average rating of 9.2.

The remaining treatments (clopyralid and hexazinone) produced only a moderate necrosis response, recording a 5.5 and 2.3 necrosis rating at 76 WAT respectively (Fig. 5).

Plant mortality

At 32 and 76 WAT, significant plant mortality differences ($P < 0.001$) were recorded between treatments, consistent with those for necrosis at the same time periods (Table 2). Mortality was minimal in the control, averaging only 0.4% at the end of the trial. At 32 WAT, mortality among metsulfuron-methyl, glyphosate and aminopyralid + metsulfuron-methyl was not significantly different ($P > 0.05$), averaging 87.1%, 67.1% and 63.8% mortality respectively. In contrast, mortality in clopyralid, hexazinone and triclopyr + picloram was not significantly different ($P > 0.05$) from the control (≤0.4%; Table 2).

By 76 WAT, aminopyralid + metsulfuron-methyl, glyphosate, metsulfuron-methyl and triclopyr + picloram treatments all recorded high mortality (≥85%), and did not significantly differ ($P > 0.05$). Although mortality had increased, clopyralid was far less effective, averaging only 32.5%. Mortality of the hexazinone treatment remained low (14.4%), and was not significantly different ($P > 0.05$) from the control at 76 WAT (Table 2).

Discussion

The Injecta® herbicide-capsule delivery method was an effective control strategy for the management of *C. uruguayanus*. The plants were able to dissolve and absorb the granular herbicides from the capsules, but efficacy varied with the herbicide applied. Glyphosate provided the most rapid plant necrosis, leading to very high mortality, a finding consistent with Campbell *et al.* (2021) who reported 91% and 99% mortality of *C. uruguayanus*, 13 and 20 months respectively, after stem injection of glyphosate, and also obtained high levels of mortality by using glyphosate as a cut-stump application.

Encapsulated glyphosate applied using the Injecta® system has also successfully controlled a range of woody weeds, including Chinese elm (*C. sinensis*), prickly acacia (*Vachellia nilotica* (Benth.) Kyal. & Boatwr.), leucaena (*Leucaena leucocephala* Lam. De Wit), camphor laurel (*Cinnamomum camphora* (L.) J.Presl) and privet (*Ligustrum lucidum* W.T. Aiton; Goulter *et al.* 2018; O'Brien *et al.* 2022). Conversely, it was relatively ineffective on mimosa bush (*V. farnesiana*; Limbongan *et al.* 2021).

Although slower to act than glyphosate, aminopyralid metsulfuron-methyl, metsulfuron-methyl, and triclopyr + picloram caused relatively high mortality at 76 WAT. Liquid triclopyr + picloram herbicides have been widely used for stem-injection treatments on exotic and native weeds (McKenzie *et al.* 2010). Campbell *et al.* (2021) found that a relatively new liquid product containing triclopyr + picloram + aminopyralid (Tordon™ RegrowthMaster) eventually caused high mortality of *C. uruguayanus*; however, it required substantial time to see an effect. At 13, 20 and 26 months after treatment (MAT), mortality averaged 22%, 79% and 99% respectively, demonstrating a relatively similar pattern to that of triclopyr + picloram in the current study.

Clopyralid and hexazinone failed to control *C. uruguayanus*. The results for hexazinone contrast with those of Campbell *et al.* (2021) who found that stem injection of liquid hexazinone caused 90% mortality 11 months after treatment, compared with only 14.4% at 76 WAT in the current study. This differential response may be associated with differences in the herbicide concentration applied. A single application (i.e. one capsule per plant) equates to 265.5 mg of hexazinone in the current study, compared with 500 mg in Campbell *et al.* (2021). Nevertheless, where *C. uruguayanus* is growing near native vegetation, alternatives to hexazinone may be preferable. Even though the stem-implantation technique contains the herbicide in the plant, herbicide could potentially leach into surrounding soil and deleteriously affect desirable plants. In such situations, less persistent herbicides would be a preferred option. Vitelli and Madigan (2011), using the Ez-Ject® herbicide lance found that despite the herbicide capsules (containing imazapyr) being directly injected into the target weed, some non-target damage occurred to native vegetation in close proximity.

Other herbicides capable of controlling *C. uruguayanus* using stem injection include amitrole + ammonium thiocyanate and MSMA (Campbell *et al.* 2021). However, these herbicides are currently unavailable in a dry formulation that would enable encapsulation using the current system. MSMA is by far the fastest-acting herbicide for *C. uruguayanus* control, but with sufficient time, other less efficacious herbicides with shorter withholding periods and lower mammal toxicity give similar levels of efficacy and would be preferable (Campbell *et al.* 2021). In contrast, amitrole + ammonium thiocyanate was one of the slowest to act, recording only 66% mortality 20 months after treatment, but eventually this increased to 96% at 36 MAT (Campbell *et al.* 2021).

Table 2. *Cereus uruguayanus* mortality (%) 32 and 76 weeks after herbicide applications.

Treatment	32 WAT	76 WAT
Control	0.0b	0.4c
Aminopyralid + metsulfuron-methyl	63.8a	97.5a
Clopyralid	0.0b	32.5b
Glyphosate	67.1b	98.3a
Hexazinone	0.4b	14.4bc
Metsulfuron-methyl	87.1a	100.0a
Triclopyr + picloram	2.5b	85.5a

Means within a column that do not share the same letter are significantly different (at $P = 0.05$) according to Tukey's HSD test.

Overall, current and past research suggests that *C. uruguayanus* is slow to respond to herbicide applications. Consequently, sufficient time should be allowed before evaluating effectiveness. The current study was undertaken during a prolonged dry period. Herbicide efficacy can reduce under such conditions because plant translocation and respiration decline, restricting herbicide movement (Parker 2005; Zhou *et al.* 2007; Department of Primary Industries and Regional Development 2021). Although the dry conditions did not appear to affect results, it is possible that under better rainfall conditions, several herbicides may have acted more rapidly.

The Injecta® herbicide-capsule delivery method keeps the plant intact throughout the treatment period, with the entire plant being exposed to the herbicide. During the Injecta® trial, any loss of branches or stems was a result of plant necrosis; no regrowth of fallen branches or stems was observed. Campbell *et al.* (2021) recorded regrowth of fallen stems in cut-stump treatments of *C. uruguayanus*, although regrowth was rare. Spray applications to the cut section of the stems increased mortality but only in small plants. They observed that *C. uruguayanus* maintained its structure when it falls, unlike other cactus species where cladodes and stem sections readily break off.

The Injecta® herbicide-capsule delivery method appears as effective as traditional stem-injection approaches for *C. uruguayanus* control. Individual plant treatment is rapid, with the advantage, from a non-target perspective, of herbicide containment within the stem. Additionally, treatment with capsules is a lightweight and convenient system, with reduced need for personal protective equipment (PPE) and minimal herbicide loss through environmental escape and un-used tank mixes. Further research should focus on quantifying these aspects by comparing cost effectiveness and the ability to minimise non-target damage against other herbicide application techniques, including other stem-injection options.

Conclusions

In this trial, *C. uruguayanus* was successfully controlled using the Injecta® herbicide-capsule delivery method. The equipment effectively implanted capsules into the stem of *C. uruguayanus*, facilitating herbicide movement throughout the plant. Of the herbicides evaluated, glyphosate, aminopyralid metsulfuron-methyl, metsulfuron-methyl and triclopyr + picloram where the most effective for control. The remaining herbicides evaluated were ineffective for control; however, for hexazinone this may be a consequence of plants receiving a non-lethal dose. The Injecta® herbicide-capsule delivery method was an efficient and effective technique for the management of cacti, particularly considering the remoteness of the trial location, the rough terrain and proximity to native vegetation.

Further studies evaluating physiological interaction of *C. uruguayanus* with the herbicides implanted using the Injecta® herbicide-capsule delivery method may provide insight regarding differences in herbicide efficacy and speed of plant mortality. Additionally, a cost analysis of this system against other control techniques, particularly those used for stem-injection applications, would be beneficial for land management of *C. uruguayanus*.

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Data availability. The data that support this study will be shared upon reasonable request to the corresponding author.

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Author affiliations

[A]Queensland Department of Agriculture and Fisheries, Gatton, Qld, Australia.

[B]School of Agriculture and Food Sciences, The University of Queensland, Gatton, Qld, Australia.