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Project Code	
Project Type	

## FINAL REPORT 2019

<b>PROJECT CODE</b> : PIR116
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### PROJECT TITLE

Silverleaf nightshade – long-term management effects of annual herbicide applications

### PROJECT DURATION

<b>Project Start date</b>	July 1 2016				
<b>Project End date</b>	June 30 2019				
<b>SAGIT Funding Request</b>	2016/17		2017/18		2018/19

### PROJECT SUPERVISOR CONTACT DETAILS

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## PROJECT REPORT

### Executive Summary

This report summarises five seasons of research on silverleaf nightshade (SLN), to early 2019. Field experiments were established near Keith and Warnertown to measure: 1) long-term effect of successive annual herbicide treatments used in cropping systems on SLN density (broad-acre trials); 2) long-term effects of “spot-spraying” herbicides (spot-spray trials); and 3) the rate of density decline under intensive glyphosate applications (large plot eradication trials).

Broad-acre experiments: Glyphosate at 3 L Prod. ha<sup>-1</sup> (540g/L) reduced SLN density at both sites but 1.5 L ha<sup>-1</sup> did not, highlighting the importance of using a robust rate. Pulse added at 0.2% improved glyphosate efficacy and should be used routinely against SLN. Glyphosate at 3 L Prod. ha<sup>-1</sup> (540g/L), with added Pulse (0.2%), appears to be the most suitable treatment for SLN, repeated in late summer/autumn if seasonal conditions allow. Starane Advanced (333 g L<sup>-1</sup> fluroxypyr) at 600 ml Prod. ha<sup>-1</sup> gave useful long-term suppression and is an effective alternative to 2,4-D amine.

Spot-spray experiments: Uragan (bromacil) and Graslan (8 kg ha<sup>-1</sup>) were the most effective. Arsenal, FallowBoss Tordon, Graslan (4 kg Prod. ha<sup>-1</sup>), Hotshot and Velmac G also have potential.

Eradication herbicide experiments: Three years after the last of three successive years of treatment with glyphosate (1.62 kg a.i. ha<sup>-1</sup>), SLN density was reduced by 90% at Warnertown and 83% at Keith.

### Project Objectives

This project (PIR116) extended, by three years, the data collection period for field herbicide experiments established in PIRSA 0113 (2013-2016) which, in part:

“Established two regional collaborative research and demonstration focus sites to involve SA grain growers and promote ownership and rapid adoption of current and newly-discovered information on SLN management.”

This project has built on existing results from PIRSA 0113 to capture data over a total of six consecutive summers following three consecutive annual herbicide treatments. It is rare that farmers are provided with long-term data on crop management, but results over this time span are especially valuable for assessing the cumulative herbicide effect and economic returns from perennial weed management.

## Overall Performance

This project has successfully achieved its goals (Jan 2019), except for delivery of two regional farmer's meetings that will be delivered by PIRSA Biosecurity SA staff in late summer/autumn 2019. The field sites were successfully established at Warnertown (with thanks to property owner Mr. Wayne Young) and at Keith (with thanks to property owners Mr. Alf Densley, Mr. John Gould, and Mr. Graham Parker) in early 2014. Treatments were successfully applied according to plan, except that late treatments in 2016 were withheld at both sites due to SLN shoots being severely droughted. Shoot density was captured annually as planned at each site. A series of farmer SLN update meetings have already been convened at various locations in SA, and two more are planned for early 2019. The experiments have identified several effective herbicide treatments for both broad-acre and spot-spraying situations, and have identified a likely time-frame (4 to 5 years) for moving from broad-acre treatments to spot-treatments. The value of collecting data over six successive seasons has been demonstrated, because significant effects could often only be detected four to five years into the research. The staff working on this project included Dr. John Heap (Principal Investigator), Dr. Ross Meffin (Group Manager), Dr. Kathy Ophel Keller (Manager after Sept., 2018) and Dr. Jane Prider (Data collection assistance and statistical analyses).

The final year of the project was subject to negotiation and amendment (SARDI/SAGIT) to accommodate Dr. Heap's acceptance of a redundancy package effective on December 21 2018. Dr. Heap completed the field data collection before his termination, and undertook to complete the analysis (with help from Dr. Prider) and reporting (Detailed technical report and this Final Report) in early 2019.

## Key Performance Indicators (KPI)

<i>KPI</i>	<i>Achieved (Y/N)</i>	<i>If not achieved, please state reason.</i>
<b>1. 2016/17: Rep-peg, count and assess broad-acre, spot-spraying and eradication experiments at two focus sites near Keith and Pt Pirie.</b>	Y	
<b>2. 2016/17: Apply annual herbicide treatments to broad-acre and eradication experiments at two focus sites near Keith and Pt Pirie.</b>	Y	
<b>3. 2017/18: Rep-peg, count and assess broad-acre, spot-spraying and eradication</b>	Y	

<b>experiments at two focus sites near Keith and Pt Pirie.</b>		
<b>4. 2017/18: Apply annual herbicide treatments to broad-acre* and eradication experiments at two focus sites near Keith and Pt Pirie.</b>	N*	After examination of the 2017-2018 density data a decision was made that the value of the early 2019 data (final data set) would be highest if the perennial root systems were allowed to regenerate without the final herbicide application in 2018. SLN roots are capable of surviving for a long time after herbicide treatment, despite apparent death of the shoots. Shoots can thereby re-emerge several years after treatment. By withholding treatments in 2017-2018 SLN roots and shoots will be allowed a full two years of recovery, allowing long-term root damage to be more accurately indicated by shoot emergence in 2019.
<b>5. 2018/19: Rep-peg, count and assess broad-acre, spot-spraying and eradication experiments at two focus sites near Keith and Pt Pirie.</b>	Y	
<b>6. Present project research results to two farmer meetings near Keith and Pt Pirie</b>	Future	Arrangements are in place for delivery of two regional farmer's meetings that will be delivered by PIRSA Biosecurity SA staff in late summer/autumn 2019
<b>7. Develop and submit Final Report to SAGIT</b>	Y	

#### **Technical Information**

Please note that a much more detailed formal research report has been submitted as an appendix to this Final Report.

This project (PIR116) extends, by three years, the data collection period for field herbicide experiments established in PIRSA 0113.

Farmers typically apply herbicides to large, established infestations of SLN during summer, but SLN is often not the primary target. Summer spraying of paddocks is usually undertaken after significant summer rainfall events, primarily to conserve soil moisture and a manageable seedbed for the following crop. Often summer weeds such as caltrop, heliotrope and melons are the primary target, and SLN is sprayed incidentally. Herbicide mixtures often reflect these priorities, and costs are reduced by using just enough glyphosate (typically around 650 g a.i. ha<sup>-1</sup>) and partner herbicides to control the annual weeds and kill the SLN shoots. Glyphosate and 2,4-D amine are most often used when SLN

is specifically-targeted. These results, collected over six years, confirm that SLN management with herbicides remains a long-term undertaking. The results illustrate a slow revelation of significant progress, which supports the value of conducting these experiments over an extended period.

The beginning of the field experiments in early January 2014 unfortunately coincided with a near record heat wave at both sites (Jan 7 to Jan 13), with daily maximum temperatures between 42 to 46°C. This stressed SLN severely, and resulted in delayed spraying at both sites to allow partial recovery. This situation was repeated in early 2015 when, despite a large rainfall event at both sites in mid-January, SLN plants were again stressed by at least five weeks without significant rainfall in late January and February. It is likely that SLN was under moderate moisture stress at both sites at the time of spraying in early 2014 and early 2015, probably hindering herbicide absorption, translocation and efficacy.

After examination of the 2017-2018 density data a decision was made that the value of the early 2019 data (final data set) would be highest if the perennial root systems were allowed to regenerate without the final herbicide application in 2018. SLN roots are capable of surviving for a long time after herbicide treatment, despite apparent death of the shoots. Shoots can thereby re-emerge several years after treatment. By withholding treatments in 2017-2018 SLN roots and shoots will be allowed a full two years of recovery, allowing long-term root damage to be more accurately indicated by shoot emergence in 2019.

There were large year to year density fluctuations in untreated control plots at both sites. The patterns were similar at both sites, reflecting general seasonal conditions in South Australia. The fluctuations were probably driven primarily by seasonal changes to available soil moisture, and the patterns seen in untreated plots were reflected in most of the treatments at both sites. Unfortunately these inherent density fluctuations have a confounding effect on statistical analyses. The trial sites chosen had a reasonably even distribution of SLN, and there were no significant differences between plots at the beginning of the experiments at either site. Keith had a higher initial density ( $8.0 \text{ shoots m}^{-2} \pm 0.3$ ) than Warnertown ( $2.3 \text{ shoots m}^{-2} \pm 0.2$ ).

In summary, the most successful treatments at Keith were glyphosate at  $3 \text{ L ha}^{-1}$ ; glyphosate at  $3 \text{ L ha}^{-1} + \text{EDTA}$ ; glyphosate at  $1.5 \text{ L} + \text{Starane at } 0.45 \text{ L ha}^{-1}$ ; glyphosate at  $1.5 \text{ L} + 2,4\text{-D amine at } 0.75 \text{ L ha}^{-1}$ ; and glyphosate at  $1.5 \text{ L}$  early then  $3 \text{ L ha}^{-1}$  late. The most successful at Warnertown were glyphosate at  $3 \text{ L ha}^{-1}$ , glyphosate at  $1.5 \text{ L ha}^{-1}$ , Starane at  $0.6 \text{ L ha}^{-1}$ , and glyphosate at  $1.5 \text{ L}$  early then  $3 \text{ L ha}^{-1}$  late. Glyphosate at  $3 \text{ L ha}^{-1}$ , and glyphosate at  $1.5 \text{ L}$  early then  $3 \text{ L ha}^{-1}$  late were the only two to perform well at both sites.

**Glyphosate** performed better at Keith than at Warnertown, probably due to higher soil water availability to SLN roots, thus increasing herbicide translocation. Nufarm Glyphosate 540 alone at  $1.5 \text{ L Prod. ha}^{-1}$  was insufficient to control SLN at either site however, with added Pulse penetrant (0.2%), it performed much better and reduced SLN density at both sites, to a level similar to  $3.0 \text{ L Prod. ha}^{-1}$  rate of glyphosate without Pulse. The  $3.0 \text{ L Prod. ha}^{-1}$  rate of glyphosate performed far better than the  $1.5 \text{ L Prod. ha}^{-1}$  rate at both sites, suggesting that the extra investment (c.  $\$8 \text{ ha}^{-1}$ ) is warranted for long-term control. Glyphosate at  $3 \text{ L Prod. ha}^{-1}$  (540g/L), with added Pulse (0.2%), appears to be the most suitable treatment for SLN, repeated in late summer/autumn if seasonal conditions allow.

The addition of **EDTA** to **glyphosate** was a speculative treatment, based on published reports of high concentrations of  $\text{Ca}^{++}$  ions on the leaf surface of some plant species, and speculation that this might “lock up” some glyphosate before it can be absorbed into the target plant. EDTA is known to bind strongly to  $\text{Ca}^{++}$  ions, and so was chosen in attempt to bind  $\text{Ca}^{++}$  ions on the SLN leaf surface, thus allowing more glyphosate to be absorbed. A concentration of 10 mM EDTA was chosen for the experiment, but Nufarm Glyphosate 540 at 3.0 L Prod. ha<sup>-1</sup> mixed with EDTA performed no better than 3.0 L Prod. ha<sup>-1</sup> alone at either site.

**Starane** (fluroxypyr) at 600 ml Prod. ha<sup>-1</sup> gave a similar level of shoot density reduction to Nufarm Glyphosate 540 at 1.5 L Prod. ha<sup>-1</sup> with added Pulse, at both sites. It performed relatively better at Warnertown than at Keith, suggesting that fluroxypyr may be more robust than glyphosate under dry conditions. Starane at 600 ml Prod. ha<sup>-1</sup> was more effective than Amicide Advanced (2,4-D amine) at 1.5 L Prod. ha<sup>-1</sup> at Warnertown, but not Keith. None of these trends were statistically significant. Given that the cost of the two treatments is similar, Starane appears to be a better choice than the long-standing treatment of 2,4-D when SLN is water-stressed. At Keith, where soil moisture is usually higher than at more northerly and westerly regions of SA, glyphosate appears to be more efficacious than Starane, and Starane and 2,4-D gave similar results. This is probably because both glyphosate and 2,4-D are translocated better through SLN roots when soil moisture is higher.

Mixtures of **glyphosate** and **Starane**, and **glyphosate** and **2,4-D amine**, appeared to synergistic at Keith, but antagonistic at Warnertown. At Keith the reduced rates of both Starane and 2,4-D amine, when mixed with Nufarm Glyphosate 540 at 1.5 L Prod. ha<sup>-1</sup>, performed better than label rates of either Starane or 2,4-D amine alone. At Warnertown both mixtures of either Starane or 2,4-D amine with glyphosate gave very poor results. These treatments should be considered with caution until more data is available, especially when SLN is under moisture stress.

**Hotshot** (aminopyralid/fluroxypyr) gave some suppression at Keith, but failed at Warnertown, and is not considered to be an effective option at this stage.

Nufarm **Glyphosate** 540 at 3.0 L Prod. ha<sup>-1</sup>, followed by **Nuquat** 250 (paraquat) the next day was not as effective as Nufarm Glyphosate 540 at 3.0 L Prod. ha<sup>-1</sup> alone. This was a speculative treatment, designed to try to overcome the problem of herbicide excretion by SLN roots. The treatment explored whether glyphosate might be introduced to the SLN root system by translocation, and then trapped there by the effective “defoliation” of the shoot, using paraquat. The concept is expensive and appears to be unsuccessful, and does not encourage further investigation.

The **split treatments** with an early summer and late summer/autumn component gave no better SLN density reduction than single early treatments alone, at either site. These included Nufarm Glyphosate 540 at 3.0 L Prod. ha<sup>-1</sup> applied in autumn, following summer applications of Nufarm Glyphosate 540 at 1.5 L Prod. ha<sup>-1</sup>, Starane at 450 ml Prod. ha<sup>-1</sup>, or Amicide Advance at 1.0 L Prod. ha<sup>-1</sup>. The combination of dry soil in autumn, and the residual root suppression from the first component of the split treatment, probably restricted the absorption and translocation, and hence efficacy, of the second glyphosate component. These results suggest that the double treatment (“Dual Action”) that is successful in NSW,

where soil moisture is higher during summer/autumn, may not be as reliable in SA or WA, or Vic.

**Garlon FallowMaster** (triclopyr) alone at 400 and 800 mL Prod. ha<sup>-1</sup> gave useful suppression at Keith, but failed at Warnertown. It performed better when mixed with glyphosate, resulting in some suppression of shoot density reduction. **Fallowboss Tordon** (picloram/aminopyralid/2,4-D amine) plus additional 2,4-D amine was ineffective.

The treatment comprising Nufarm **Glyphosate 540** at 1.2 L Prod. ha<sup>-1</sup>, plus **Garlon FallowMaster** at 80 mL Prod. ha<sup>-1</sup>, plus Associate (metsulfuron-methyl) at 7g Prod. ha<sup>-1</sup> was included to represent typical mixtures applied by farmers to control mixed populations of summer weeds (e.g. melons, heliotrope, caltrop etc.). Similar herbicide treatments are incidentally applied to SLN while spraying other summer weeds, with the expectation of at least SLN shoot desiccation. While shoot desiccation is often achieved, the results from both sites suggest that little long-term damage is done to SLN roots.

These experiments have focused on the long-term reduction in SLN shoot density, relying mostly on annual density data from permanent quadrats. Visual effects on SLN shoots several weeks after treatment are variable, and sometimes confounded by drought symptoms. Two reliable sets of data on SLN biomass reduction (visual assessment of % live biomass reduction), 17 and 26 days after herbicide treatments (DAT), were collected at Warnertown in March 2014 and February 2015. These results show that high levels of shoot damage, in the weeks after treatment, do not necessarily correlate with reductions in shoot density. Conditions before and after spraying in 2015 were drier than those in 2014. Glyphosate, in particular, resulted in only slight to moderate visible shoot damage at 17 and 26 DAT. Starane gave excellent shoot control and also reduced shoot density over time. Starane and 2,4-D mixtures with glyphosate also controlled shoot growth quickly. Garlon FallowMaster (triclopyr), Fallowboss Tordon plus additional 2,4-D amine, and the glyphosate Garlon FallowMaster plus Associate mixture all demonstrated effective short term shoot on at least one occasion.

An apparent increase in SLN shoot density following herbicide application was sometimes observed, especially at Warnertown. This phenomenon probably results from the isolation of segments of the root system, and subsequent growth of buds released from the suppressive effects of apical dominance. This phenomenon has important implications for both farmer's perceptions and long-term SLN control tactics. Firstly, if farmers are unaware of the likely mechanism behind the apparent increase in shoot density, they may conclude that the herbicide has made matters worse and discontinue treatments. More seriously, in some situations herbicides may indeed increase the SLN shoot density if daughter shoots from isolated root fragments are not controlled. This potential situation was observed at Keith in autumn, 2016. Two successive applications of 1620 g.a.i. ha<sup>-1</sup> had led to a rapid decline in shoot density by February 2016. However, 90 mm of subsequent rainfall produced a dense cohort of young, small SLN shoots in the treated plots, but not in the untreated. Given the high levels of soil moisture present, it would be very likely that these shoots would develop to become autonomous established perennial plants. Thus, beginning and then abandoning long-term management using glyphosate may increase the SLN density, leading to higher yield losses. Farmers should be warned that, once management commences, it is important to continue until carbohydrate reserves are exhausted in the isolated sections of the root system, so that adventitious buds can no longer reach the soil surface.

## Conclusions Reached &/or Discoveries Made

The following discoveries and conclusions have been made:

- 1) Field research on control of deep-rooted perennial weeds provides data that are more reliable after six years than after only three years.
- 2) In eradication experiments, three years after the third season of successive applications of glyphosate (1.62 kg a.i. ha<sup>-1</sup>), density was reduced by 90% at Warnertown and 83% at Keith. It appears possible that long-term use of broad-acre herbicide applications could reduce shoot density to a point where spot spraying is feasible.
- 3) Reduction of shoot density requires a persistent long-term commitment, using robust herbicide application rates.
- 4) Glyphosate at 3 L Prod. ha<sup>-1</sup> (540g/L), with added Pulse (0.2%), appears to be the most suitable treatment for SLN, repeated in late summer/autumn if seasonal conditions allow.
- 5) Glyphosate at 1.62 kg a.i. ha<sup>-1</sup> is more effective than 0.81 kg a.i. ha<sup>-1</sup> in the long-term, despite the lower rate killing shoots in the short-term.
- 6) Pulse penetrant increased the efficacy of glyphosate and should be considered as a standard adjuvant with SLN.
- 7) Starane Advanced (fluroxypyr) at 200 g a.i. ha<sup>-1</sup> has the potential to reduce shoot density over the long-term, and is as effective as 2,4-D amine.
- 8) The “Dual-Action” (e.g. early and late spray) strategy employed in NSW may not be as effective in SA because plants may not recover from the early treatment due to lack of rain.
- 9) Uragan and Graslan (8 kg ha<sup>-1</sup>) were the most effective spot spraying treatments. Arsenal, FallowBoss Tordon, Graslan (4 kg Prod. ha<sup>-1</sup>), Hotshot and Velmac G also have potential.

## Intellectual Property

None identified.

## Application / Communication of Results

The major findings of this project are presented in dot-point form in the Conclusions/Discovery section above.

Industry significance. These findings will provide growers with more reliable information to manage SLN in cropping rotations with more confidence. There was already evidence that controlling SLN shoots during summer increased the following crop yield. This research has identified the importance of using higher rates of herbicides (glyphosate, fluroxypyr) to reduce shoot density over four to five years, in addition to increasing crop yield. It has also been identified that fluroxypyr reduces shoot density in the long-term, and that it is a good alternative to 2,4-D amine. This information can be adopted by increasing the glyphosate rate to 1.62 kg a.i. ha<sup>-1</sup> (using 0.2% Pulse) in summer weed control “brews”, or incorporating fluroxypyr where appropriate. Shoot density reductions of 83 to 90% recorded after six years will encourage growers to persist with long-term programs, even though the shoot density reduction may not be apparent for three or so years. The research



has also confirmed Graslan (tebuthiuron) as an effective spot treatment for eradication of small patches, and identified Uragan (bromacil) as an effective spot treatment.

Extension. The interim results from this research were presented to a number of farmer/public meetings and conferences over the last two years, including the 20<sup>th</sup> Australasian Weed Conference (Sept., 2016), AMLR NRM seminars (May, 2017 and Feb, 2018), Yackamoorundie LandCare Group (Aug, 2017), SAGIT Board Meeting (Spring, 2018), Farmer meeting at Mt Pleasant (March, 2018), Spalding farmer meeting (March, 2018), UniSA NRM weeds lecture (Aug, 2018), and the SA Biennial Weeds Conference. In addition, presentation of the results to farmer meetings in Keith and Warnertown is planned for late summer/autumn 2019, based on these results and the publication of the SLN management manual discussed below.

Path to market: *Comprehensive SLN management manual.* The “Australian SLN best practice management manual 2018” was published in September 2018, and is currently being distributed as the major path to market for this research. It was written by Dr. John Heap (SARDI) and Dr. Hanwen Wu (NSW DPI), using information generated and collected by the two SLN researchers over several decades. A box of the manuals was delivered to SAGIT in Oct, 2018. Much of the research underpinning the manual was funded by SAGIT (SA) and MLA (NSW). It is a comprehensive manual, dealing with a wide range of information on SLN biology and management, which is expected to be the major publication on SLN for at least 10 years.

## **POSSIBLE FUTURE WORK**

There are a number of potential SLN research projects that may warrant future funding:

- 1) The optimum rate of Pulse penetrant used with glyphosate against SLN.
- 2) The optimum rate of fluroxypyr for maximum SLN root damage.
- 3) Seek a permit for use of Graslan granules for spot treatment of SLN in SA.
- 4) Define the situational suitability for use of Uragan (bromacil) as a spot treatment.
- 5) Manipulation of SLN leaf surface structure (i.e. waxy branched trichome hairs) and chemistry (e.g.  $Ca^{++}$ ), including leaf surface saturation, to improve herbicide absorption.
- 6) Survey paddock infestations to determine whether SLN spatial distribution is correlated with patches of hostile sub-soil. This has implications for the economic importance of SLN in some situations.

# *Silverleaf nightshade – long-term management effects of annual herbicide applications*

## **Final Detailed Report**

**Dr John Heap, January 2019**

### **Abstract**

This report summarises five seasons of research on silverleaf nightshade (SLN), to early 2019. Field experiments were established near Keith and Warnertown to measure: 1) long-term effect of successive annual herbicide treatments used in cropping systems on SLN density (broad-acre trials); 2) long-term effects of “spot-spraying” herbicides (spot-spray trials); and 3) the rate of density decline under intensive glyphosate applications (large plot eradication trials).

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### **Summary**

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In summary, the most successful treatments at Keith were glyphosate at  $3 \text{ L ha}^{-1}$ ; glyphosate at  $3 \text{ L ha}^{-1} + \text{EDTA}$ ; glyphosate at  $1.5 \text{ L} + \text{Starane at } 0.45 \text{ L ha}^{-1}$ ; glyphosate at  $1.5 \text{ L} + 2,4\text{-D amine at } 0.75 \text{ L ha}^{-1}$ ; and glyphosate at  $1.5 \text{ L}$  early then  $3 \text{ L ha}^{-1}$  late. The most successful at Warnertown were glyphosate at  $3 \text{ L ha}^{-1}$ , glyphosate at  $1.5 \text{ L ha}^{-1}$ , Starane at  $0.6 \text{ L ha}^{-1}$ , and glyphosate at  $1.5 \text{ L}$  early then  $3 \text{ L ha}^{-1}$  late. Glyphosate at  $3 \text{ L ha}^{-1}$ , and glyphosate at  $1.5 \text{ L}$  early then  $3 \text{ L ha}^{-1}$  late were the only two to perform well at both sites.

**Glyphosate** performed better at Keith than at Warnertown, probably due to higher soil water availability to SLN roots, thus increasing herbicide translocation. Nufarm Glyphosate 540 alone at  $1.5 \text{ L Prod. ha}^{-1}$  was insufficient to control SLN at either site however, with added Pulse penetrant (0.2%), it performed much better and reduced SLN density at both sites, to a level similar to  $3.0 \text{ L Prod. ha}^{-1}$  rate of glyphosate without Pulse. The  $3.0 \text{ L Prod. ha}^{-1}$  rate of glyphosate performed far better than the  $1.5 \text{ L Prod. ha}^{-1}$  rate at both sites, suggesting that the extra investment (c.  $\$8 \text{ ha}^{-1}$ ) is warranted for long-term control. Glyphosate at  $3 \text{ L Prod. ha}^{-1}$  (540g/L), with added Pulse (0.2%), appears to be the most suitable treatment for SLN, repeated in late summer/autumn if seasonal conditions allow.

The addition of **EDTA** to **glyphosate** was a speculative treatment, based on published reports of high concentrations of  $\text{Ca}^{++}$  ions on the leaf surface of some plant species, and speculation that this might “lock up” some glyphosate before it can be absorbed into the target plant. EDTA is known to bind strongly to  $\text{Ca}^{++}$  ions, and so was chosen in attempt to bind  $\text{Ca}^{++}$  ions on the SLN leaf surface, thus allowing more glyphosate to be absorbed. A concentration of

10 mM EDTA was chosen for the experiment, but Nufarm Glyphosate 540 at 3.0 L Prod. ha<sup>-1</sup> mixed with EDTA performed no better than 3.0 L Prod. ha<sup>-1</sup> alone at either site.

**Starane** (fluroxypyr) at 600 ml Prod. ha<sup>-1</sup> gave a similar level of shoot density reduction to Nufarm Glyphosate 540 at 1.5 L Prod. ha<sup>-1</sup> with added Pulse, at both sites. It performed relatively better at Warnertown than at Keith, suggesting that fluroxypyr may be more robust than glyphosate under dry conditions. Starane at 600 ml Prod. ha<sup>-1</sup> was more effective than Amicide Advanced (2,4-D amine) at 1.5 L Prod. ha<sup>-1</sup> at Warnertown, but not Keith. None of these trends were statistically significant. Given that the cost of the two treatments is similar, Starane appears to be a better choice than the long-standing treatment of 2,4-D when SLN is water-stressed. At Keith, where soil moisture is usually higher than at more northerly and westerly regions of SA, glyphosate appears to be more efficacious than Starane, and Starane and 2,4-D gave similar results. This is probably because both glyphosate and 2,4-D are translocated better through SLN roots when soil moisture is higher.

Mixtures of **glyphosate** and **Starane**, and **glyphosate** and **2,4-D amine**, appeared to synergistic at Keith, but antagonistic at Warnertown. At Keith the reduced rates of both Starane and 2,4-D amine, when mixed with Nufarm Glyphosate 540 at 1.5 L Prod. ha<sup>-1</sup>, performed better than label rates of either Starane or 2,4-D amine alone. At Warnertown both mixtures of either Starane or 2,4-D amine with glyphosate gave very poor results. These treatments should be considered with caution until more data is available, especially when SLN is under moisture stress.

**Hotshot** (aminopyralid/fluroxypyr) gave some suppression at Keith, but failed at Warnertown, and is not considered to be an effective option at this stage.

Nufarm **Glyphosate** 540 at 3.0 L Prod. ha<sup>-1</sup>, followed by **Nuquat** 250 (paraquat) the next day was not as effective as Nufarm Glyphosate 540 at 3.0 L Prod. ha<sup>-1</sup> alone. This was a speculative treatment, designed to try to overcome the problem of herbicide excretion by SLN roots. The treatment explored whether glyphosate might be introduced to the SLN root system by translocation, and then trapped there by the effective “defoliation” of the shoot, using paraquat. The concept is expensive and appears to be unsuccessful, and does not encourage further investigation.

The **split treatments** with an early summer and late summer/autumn component gave no better SLN density reduction than single early treatments alone, at either site. These included Nufarm Glyphosate 540 at 3.0 L Prod. ha<sup>-1</sup> applied in autumn, following summer applications of Nufarm Glyphosate 540 at 1.5 L Prod. ha<sup>-1</sup>, Starane at 450 ml Prod. ha<sup>-1</sup>, or Amicide Advance at 1.0 L Prod. ha<sup>-1</sup>. The combination of dry soil in autumn, and the residual root suppression from the first component of the split treatment, probably restricted the absorption and translocation, and hence efficacy, of the second glyphosate component. These results suggest that the double treatment (“Dual Action”) that is successful in NSW, where soil moisture is higher during summer/autumn, may not be as reliable in SA or WA, or Vic.

**Garlon FallowMaster** (triclopyr) alone at 400 and 800 mL Prod. ha<sup>-1</sup> gave useful suppression at Keith, but failed at Warnertown. It performed better when mixed with glyphosate, resulting in some suppression of shoot density reduction. **Fallowboss Tordon** (picloram/aminopyralid/2,4-D amine) plus additional 2,4-D amine was ineffective.

The treatment comprising Nufarm **Glyphosate** 540 at 1.2 L Prod. ha<sup>-1</sup>, plus **Garlon FallowMaster** at 80 mL Prod. ha<sup>-1</sup>, plus Associate (metsulfuron-methyl) at 7g Prod. ha<sup>-1</sup> was included to represent typical mixtures applied by farmers to control mixed populations of summer weeds (e.g. melons, heliotrope, caltrop etc.). Similar herbicide treatments are incidentally applied to SLN while spraying other summer weeds, with the expectation of at least SLN shoot desiccation. While shoot desiccation is often achieved, the results from both sites suggest that little long-term damage is done to SLN roots.

These experiments have focused on the long-term reduction in SLN shoot density, relying mostly on annual density data from permanent quadrats. Visual effects on SLN shoots several weeks after treatment are variable, and sometimes confounded by drought symptoms. Two reliable sets of data on SLN biomass reduction (visual assessment of % live biomass reduction), 17 and 26 days after herbicide treatments (DAT), were collected at Warnertown in March 2014 and February 2015. These results show that high levels of shoot damage, in the weeks after treatment, do not necessarily correlate with reductions in shoot density. Conditions before and after spraying in 2015 were drier than those in 2014. Glyphosate, in particular, resulted in only slight to moderate visible shoot damage at 17 and 26 DAT. Starane gave excellent shoot control and also reduced shoot density over time. Starane and 2,4-D mixtures with glyphosate also controlled shoot growth quickly. Garlon FallowMaster (triclopyr), Fallowboss Tordon plus additional 2,4-D amine, and the glyphosate Garlon FallowMaster plus Associate mixture all demonstrated effective short term shoot on at least one occasion

An apparent increase in SLN shoot density following herbicide application was sometimes observed, especially at Warnertown. This phenomenon probably results from the isolation of segments of the root system, and subsequent growth of buds released from the suppressive effects of apical dominance. This phenomenon has important implications for both farmer's perceptions and long-term SLN control tactics. Firstly, if farmers are unaware of the likely mechanism behind the apparent increase in shoot density, they may conclude that the herbicide has made matters worse and discontinue treatments. More seriously, in some situations herbicides may indeed increase the SLN shoot density if daughter shoots from isolated root fragments are not controlled. This potential situation was observed at Keith in autumn, 2016. Two successive applications of 1620 g.a.i. ha<sup>-1</sup> had led to a rapid decline in shoot density by February 2016. However, 90 mm of subsequent rainfall produced a dense cohort of young, small SLN shoots in the treated plots, but not in the untreated. Given the high levels of soil moisture present, it would be very likely that these shoots would develop to become autonomous established perennial plants. Thus, beginning and then abandoning long-term management using glyphosate may increase the SLN density, leading to higher yield losses. Farmers should be warned that, once management commences, it is important to continue until carbohydrate reserves are exhausted in the isolated sections of the root system, so that adventitious buds can no longer reach the soil surface.

The following discoveries and conclusions have been made:

- 1) Field research on control of deep-rooted perennial weeds provides data that are more reliable after six years than after only three years.
- 2) In eradication experiments, three years after the third season of successive applications of glyphosate (1.62 kg a.i. ha<sup>-1</sup>), density was reduced by 90% at Warnertown and 83% at Keith. It appears possible that long-term use of broad-acre herbicide applications could reduce shoot density to a point where spot spraying is feasible.

- 3) Reduction of shoot density requires a persistent long-term commitment, using robust herbicide application rates.
- 4) Glyphosate at 3 L Prod. ha<sup>-1</sup> (540g/L), with added Pulse (0.2%), appears to be the most suitable treatment for SLN, repeated in late summer/autumn if seasonal conditions allow.
- 5) Glyphosate at 1.62 kg a.i. ha<sup>-1</sup> is more effective than 0.81 kg a.i. ha<sup>-1</sup> in the long-term, despite the lower rate killing shoots in the short-term.
- 6) Pulse penetrant increased the efficacy of glyphosate and should be considered as a standard adjuvant with SLN.
- 7) Starane Advanced (fluroxypyr) at 200 g a.i. ha<sup>-1</sup> has the potential to reduce shoot density over the long-term, and is as effective as 2,4-D amine.
- 8) The “Dual-Action” (e.g. early and late spray) strategy employed in NSW may not be as effective in SA because plants may not recover from the early treatment due to lack of rain.
- 9) Uragan and Graslan (8 kg ha<sup>-1</sup>) were the most effective spot spraying treatments. Arsenal, FallowBoss Tordon, Graslan (4 kg Prod. ha<sup>-1</sup>), Hotshot and Velmac G also have potential.

## Introduction

International research on control of SLN with herbicides has been ongoing since the 1960s. Results from Australian research, over the same time, have confirmed that established SLN is extremely difficult to kill. Although there has been some success, notably using picloram or glyphosate, herbicides typically only suppress shoot growth. Herbicides capable of killing established plants with one application are expensive, and sterilize the soil for years. These herbicides are useful to control small infestations, but are not suited to controlling large established infestations in agricultural enterprises.

Experience and experimental results suggests that, although shoot re-growth follows most herbicide applications to SLN, there is some damage done to the root system. In this way, annual applications may be expected to slowly deplete carbohydrate stores in the root system, leading to eventual reduction in plant vigour and density.

Three experiments were established at each of two sites to measure; 1) the long-term effect of herbicide treatments suitable for use in cropping systems on SLN density (broad acre trials); 2) the long-term effects of “spot-spraying” herbicides (spot spray trials); and 3) the rate of density decline achievable under intensive glyphosate applications (large plot trials).

This report summarises five consecutive seasons of the ongoing research, to early 2019. The research was generously funded by the South Australian Grains Industry Trust (SAGIT) and conducted by PIRSA/SARDI.

## Materials and Methods

Each of these trials comprised three replicate treatment plots. The response variable, SLN density, was measured as counts of stems in multiple 1 m<sup>2</sup> quadrats within each replicate plot. SLN density was recorded before treatments were applied (initial) and in January 2015 (M1), December 2015 (M2), January 2017 (M3), December 2017 (M4) and December 2018 (M5).

Sites were chosen near Warnertown (Fig. 1; red sandy loam; UTM Zone 54; 233920 E; 6312 2720 N) in the mid-north, and near Keith (Fig. 2; white sand over clay; Zone 54: 436 210 E; 600 4920 N) in the upper south east of South Australia in 2012. The three experiments were duplicated at each site, and were situated together on dense, even stands of SLN on flat ground. A population of *Solanum coactiliferum* (western nightshade) was scattered throughout the Warnertown site (Fig 3), and some comparative data was collected for this species. Treatment dates for herbicide applications are shown in Table 1.

*Broad-acre treatments.* Treatment plots (10 x 3m) were arranged in three replicates in a randomized block design, with 1m walkways separating replicates. There were also 1m buffers between plots. Three permanent 1 x 1m quadrat positions (Fig. 4) were established in each plot, and re-established before each density assessment using a tape measure. The initial density was recorded when the experiments were established, and then again each summer, prior to any herbicide application. Herbicide effect on shoot growth was recorded 3 to 6 weeks after herbicide application, using visual estimates of biomass reduction. Treatments (Table 2) were selected on cost, and soil residues compatible with cropping rotations. In general, treatment costs were constrained below \$30 ha<sup>-1</sup> (2013) and were applied at 112 L ha<sup>-1</sup> at 200 kPa, using a 2m wide boom, 12v electric bike sprayer (Fig. 5 and 6), fitted with Airmix orange 01 air-induction nozzles, travelling at 1 metre per second.

*Spot-spraying treatments.* Treatment plots (4 x 4m) were arranged in three replicates in a randomized block design. Two permanent 1 x 1m quadrat positions were established in each plot, and re-established before each density assessment using a tape measure. The initial density was recorded when the experiments were established, and then again each season prior to herbicide application. Liquid treatments (Table 3) were applied using a Hardi 15L backpack sprayer fitted with a single high output fan nozzle delivering 1.22 L min<sup>-1</sup> at 100 kPa (regulated) over a fan width of 90 cm. Application rate was achieved by using a digital timer with an audible alarm. Granular treatments were mixed into 2 kg of fine dry sand and scattered by hand to achieve an even distribution. Large granules (e.g. Graslan) were crushed into a coarse powder before mixing with sand. Treatments were applied to cover each plot twice, the second coverage being at right angles to the first. Treatments were applied only for two consecutive seasons, then the long-term residual effects were measured.

*Glyphosate large plot experiments.* Large treatment plots (10 x 10m) were arranged in two replicates in a randomized block design, with 1m walkways separating replicates. Twelve permanent 1 x 1m quadrat positions were established in each plot, and re-established before each density assessment. The initial density was recorded when the experiments were established, and then again each season prior to herbicide applications. Herbicide effect on shoot growth was also recorded, using visual estimates of biomass reduction. The stem diameter of 30 shoots per plot was measured 20 mm above soil level. Glyphosate was applied at 1620 g a.i. ha<sup>-1</sup> plus 1% ammonium sulphate and 0.5% LI700, using the 2m boom bike sprayer described above. Treatments were applied each season (Table 1) and the larger plots, more permanent quadrats per plot, and stem diameter measurements allowed more precise measurements of SLN density decline.

### *Statistical methods*

Broad-acre trials. As previous analyses had found significant differences in SLN density at each site, the sites were analysed separately. The mean SLN density of the three quadrats in each replicate plot were used as the response variable in a repeated measures analysis of variance (rmANOVA) examining treatment and repeated measures over time and their interaction as fixed effects. Data were transformed to log-normal values prior to analysis to achieve variance homogeneity. As the sphericity assumption was not met, a Huynh-Feldt (HF) correction was used to determine degrees of freedom and adjusted p-values. The difference between initial SLN density and density at each monitoring period was also used as a response variable (untransformed values) for a further rmANOVA analysis testing treatment, repeated measures over time and their interaction.

Spot spray trials. The SLN density counts for the two quadrats within each replicate plot were averaged. Spare untreated control plots were removed from the data set. Treatments with zero values across all replicates following herbicide application were not included in the analysis. A Generalised Linear Mixed Model (GLMM) was fitted to the plant density data for all of the monitoring times. Treatment was a fixed effect and monitoring time was a random effect in these models. Negative binomial GLMMs were fitted with a logarithmic link function. Models were fitted with and without the fixed effect (treatment) and each model compared with Chi-square tests. R software Ver. 3.3.2 was used for the analysis.

Eradication trials. The total SLN density of the 12 quadrats in each plot were used as the response variable in a repeated measures analysis of variance (rmANOVA) using the same



method as the broad acre trial. Sites were analysed separately as initial SLN densities were markedly different. These models tested the effects of treatment, time and their interaction on SLN density. The univariate degrees of freedom within models were corrected using the Greenhouse-Geissner epsilom estimate as a more conservative estimate of p-values to account for sphericity.

SLN density was recorded before treatments were applied (initial) in January 2014, and in January 2015 (M1), December 2015 (M2), January 2017 (M3), December 2017 (M4), and December 2018(M5). Following model fitting for each trial, Tukey HSD tests were used for *post hoc* multiple comparisons with significance values  $\alpha < 0.05$ . The software package JMP (Ver. 14.1.0) was used for rmANOVA analyses.



Figure 1. Warnertown herbicide research site.



Figure 2. Keith herbicide research site



Figure 3. *Solanum coactiliferum* (western nightshade) (L) co-mingled with SLN (R).



Figure 5. Plot sprayer (2m; 12 volt motor) used to apply broad-acre herbicides.



Figure 4. Tapes and quadrat used to monitor permanent quadrat positions.



Figure 6. Plot sprayed with 2m wide sprayer, showing residual damage to *Medicago* spp. in autumn (Warnertown).

Table 1. Herbicide treatment dates at Warnertown and Keith sites.

Treatment	Warnertown	Keith
2014 early	18/02/14	25/02/14
2014 late	04/04/14	06/04/14
2015 early	19/01/15	29/01/15
2015 late	14/04/15	03/03/15
2016 early	10/03/16	24/03/16
2016 late	Not applied – SLN water stressed and withered at both sites	
2017 to 2018	Monitor long-term	Monitor long-term
<b>Spot-spraying experiments</b>		
2014	06/03/14	26/02/14
2015	22/01/15	03/03/15
2016 to 2018	Monitor long-term	Monitor long-term
<b>Glyphosate large plot experiments</b>		
2014	06/03/14	13/03/14
2015	18/01/15	29/01/15
2016	09/03/16	24/03/16
2017 to 2018	Monitor long-term	Monitor long-term

Table 2. Broad-acre cropping herbicide treatments applied at Warnertown and Keith sites in three consecutive seasons.

	Herbicide ®	Active	Formulation	Rate ha <sup>-1</sup>	Cost ha <sup>-1</sup> *	Adjuvants**
1	Nufarm Glyphosate 540	glyphosate	540 g/L	1.5 L	(\$8)	1% AS + LI 0.5%
2	Nufarm Glyphosate 540	glyphosate	540 g/L	3.0 L	(\$16)	1% AS + LI 0.5%
3	Nufarm Glyphosate 540 + EDTA	glyphosate + EDTA	540 g/L	3.0 L + 417 g	(>\$16)	EDTA (10mM) + LI 0.5%
4	Nufarm Glyphosate 540	Glyphosate + Pulse	540 g/L	1.5 L	(\$8)	0.2% Pulse
5	Starane Advanced	fluroxypyr	333 g/L	600 ml	(\$13)	1% Uptake oil
6	Amicide Advance	2,4-D amine	700 g/L	1.5 L	(\$14)	LI 0.5%
7	Nufarm Glyphosate 540 + Starane Advanced	glyphosate + fluroxypyr	540 g/L + 333 g/L	1.5 L + 450 ml	(\$18)	1% AS + 1% Uptake oil
8	Nufarm Glyphosate 540 + Amicide Advance	glyphosate + 2,4-D amine	540 g/L + 700 g/L	1.5 L + 750 ml	(\$15)	1% AS
9	Hotshot	aminopyralid/ fluroxypyr	10 g/L/ 140 g/L	500 ml	(\$19)	-
10	Nufarm Glyphosate 540 then Nuquat 250 1 DAT (glypho)	glyphosate then paraquat	540 g/L then 250 g/L	3.0 L then 3.0 L	(\$43)	1% AS + LI 0.5%
11	Nufarm Glyphosate 540 early then again in March	glyphosate + glyphosate	540 g/L	1.5 L then 3.0 L	(\$24)	1% AS + LI 0.5%
12	Starane Advanced early then Nufarm Glyphosate 540 in March	fluroxypyr then glyphosate	333 g/L then 540 g/L	450 ml then 3.0 L March	(\$26)	1% Uptake oil then 1% AS + LI 0.5%
13	Amicide Advance early then Nufarm Glyphosate 540 in March	2,4-D amine then glyphosate	700 g/L then 540 g/L	1.0 L then 3.0 L	(\$25)	LI 0.5% then 1% AS + LI 0.5%
14	Garlon FallowMaster	triclopyr	755 g/L	400 ml	(\$9)	0.5% Uptake oil
15	Garlon FallowMaster	triclopyr	755 g/L	800 ml	(\$18)	0.5% Uptake oil
16	FallowBoss Tordon + Amicide Advance	2,4-D amine/ picloram/ aminopyralid + 2,4-D amine	300/75/7.5 g/L + 700 g/L	300 ml + 330 ml	(\$13)	LI 0.5%
17	Nufarm Glyphosate 540 + Garlon FallowMaster + Associate	glyphosate + triclopyr + metsulfuron	540 + 755 g/L + 600g/kg	1.2L + 80ml + 7g	(\$9)	1% AS + LI 0.5%
18	Garlon FallowMaster + Nufarm Glyphosate 540	triclopyr + glyphosate	755 + 540 g/L	400 ml + 1.5 L	(\$19)	0.5% Uptake oil
19	Spare plots	-	-	-	-	-
20	Untreated Control	-	-	-	-	-

\*Approximate cost of herbicides in 2013 – check current prices from suppliers.

\*\*AS = ammonium sulphate (applied as Liaise®); LI= LI 700® adjuvant

Table 3. Spot-spraying herbicide treatments applied at Warnertown and Keith sites in three consecutive seasons.

Treat No.	Herbicide ®	Active	Form	Rate	Adjuvants
1	Graslan	tebuthiuron	200g/kg	4 kg/ha	Spread dry
2	Graslan	tebuthiuron	200g/kg	8 kg/ha	Spread dry
3	FallowBoss Tordon	2,4-D amine/ picloram/aminopyralid	300/75/ 7.5 g/L	15 L/ha	-
4	Nufarm Glyphosate 540	glyphosate	540 g/L	1:100	1% SA
5	Nufarm Glyphosate 540 + EDTA	glyphosate + EDTA	540 g/L	1:100 + 3.72g per L water	EDTA (10mM) + LI 0.5%
6	Garlon FallowMaster	triclopyr	755 g/L	400 ml /100L	-
7	Hotshot	aminopyralid/fluroxypyr	10 /140 g/L	5 L	-
8	Hotshot	aminopyralid/fluroxypyr	10 /140 g/L	10 L	-
9	Trimac	terbacil/sulfometuron	880/40 g/kg	1 kg/ha	Spread dry
10	Velmac G	hexazinone	200 g/kg	19 kg/ha	Spread dry
11	Arsenal Xpress	imazapyr/glyphosate	150/150 g/L	10 L/ha	0.2% Pulse
12	Uragan	bromacil	800 g/kg	20 kg/ha	Spread dry
13	Untreated Control	-	-	-	-

\*AS = ammonium sulphate (applied as Liaise®); LI= LI 700® adjuvant.

## Results

### *Broad-acre trials*

#### *Keith*

Initially there was no difference in SLN density among plots (*ANOVA*  $F = 1.03$ ,  $df = 18$ ,  $P = 0.45$ ). There was an average of  $8 \pm 0.3$  SLN plants  $m^{-2}$ . Although there was no overall significant difference between herbicide treatments, there were significant differences between the treatments at different monitoring times (time by treatment effect, Table 4). The treatment by time interaction indicates differences in the slope of the responses over time. It was predicted that in control plots the density of SLN would remain relatively constant whereas in herbicide treated plots it would decline over time if the herbicides successfully controlled SLN. Multiple comparison tests (Tukey HSD tests) lacked power to detect differences between monitoring periods for each herbicide treatment (see Fig. 7), therefore the difference between initial SLN density and density at each monitoring period was analysed. In this analysis the time by treatment effect was not significant, indicating there were no significant further changes to SLN density at each subsequent monitoring time (Table 5).

Table 4. Results of rmANOVA testing the effects of site and herbicide treatment on SLN density over time in the broad acre trial at Keith.

Effects	Value	Num DF	Den DF	Exact F	Pr > F
Treatment	0.02	1	35	1.773	0.35
Time	0.82	4.08	224.6	18.62	< 0.001
Treatment*Time	0.82	4.08	224.6	2.38	0.05

Table 5. Results of rmANOVA testing the effects of site and herbicide treatment on changes in initial SLN density over time in the broad acre trial at Keith.

Effects	Value	Num DF	Den DF	Exact F	Pr > F
Treatment	0.22	1	55	11.84	0.001
Time	0.40	4	52	5.14	0.002
Treatment*Time	0.06	4	52	0.80	0.53

There was a significant difference between the changes in SLN density at each monitoring time (time effect, Table 5). There was only a minor decrease in SLN density at M3, less than 1 plant  $m^{-2}$  compared to initial density (Fig. 8). At all other monitoring times there was on average 4-5 fewer plants  $m^{-2}$  than initially.

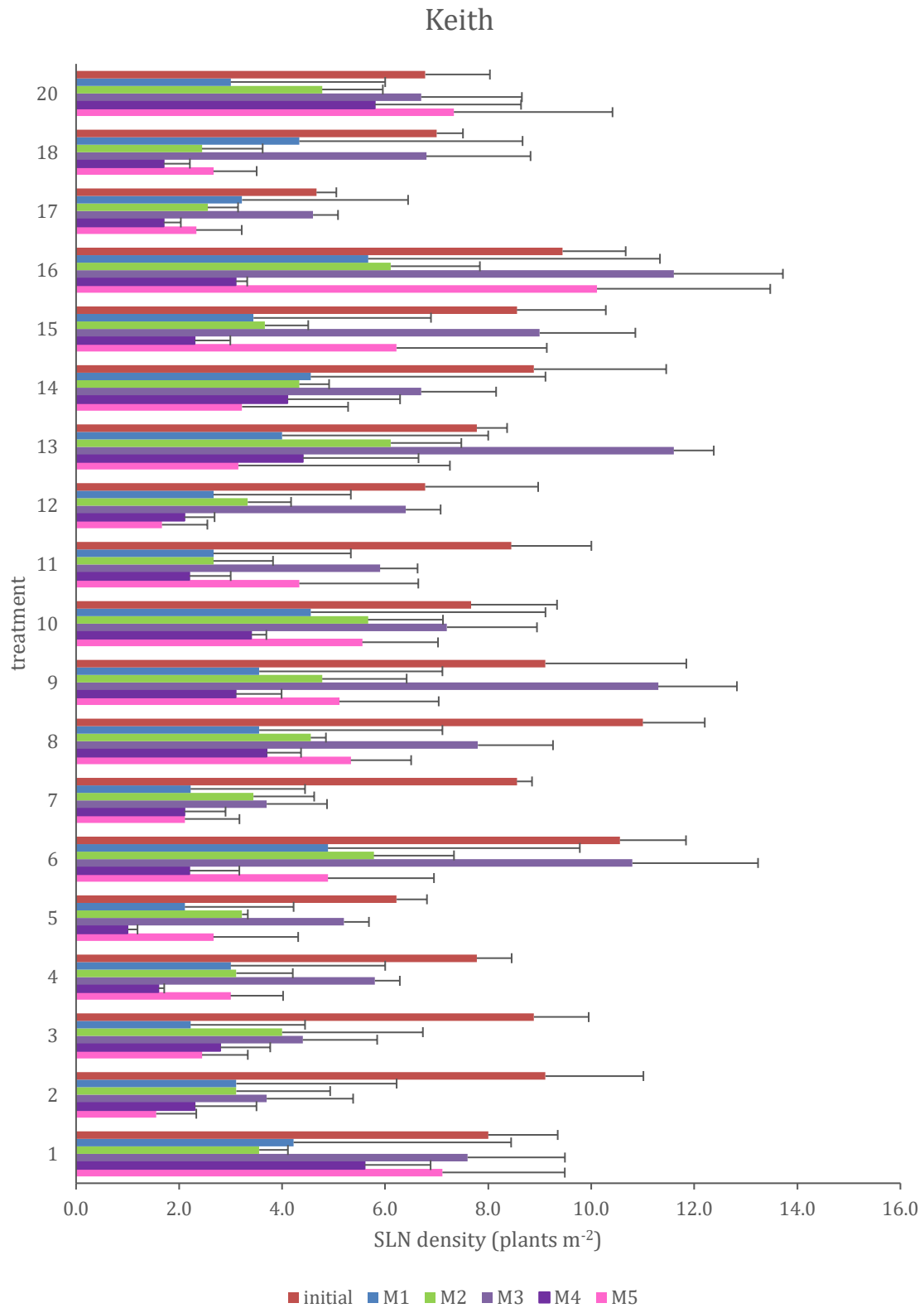


Figure 7. SLN density (mean + 1 SE) in the broad-acre trial before herbicides were applied (initial) and at five successive annual monitoring times at the Keith site. Untreated controls are treatment 20. See Table 2 above for identity of other treatments.

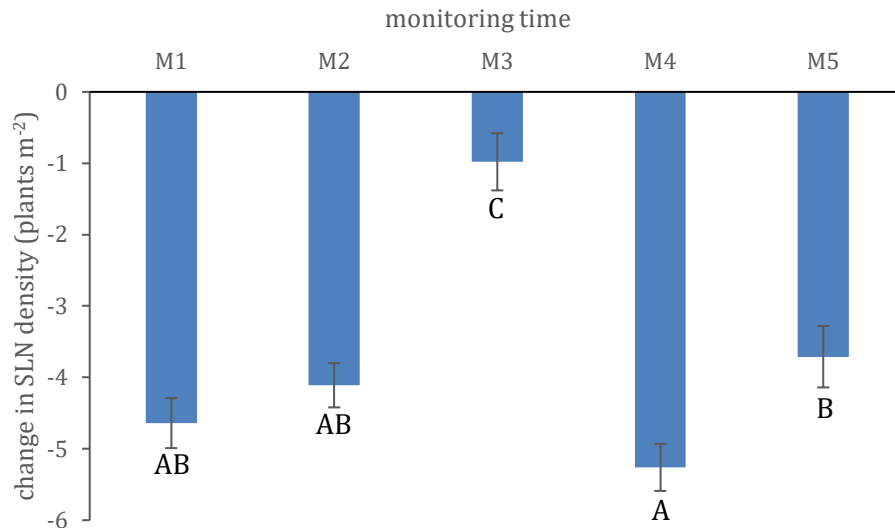


Figure 8. Changes in SLN plant density in the broad-acre trial at Keith over the monitoring period (treatments combined). Bars labelled with a different letter were significantly different ( $p < 0.05$ ). Bars are means + 1SE.

Overall, treatments 2, 3, 7, 8 and 11 had significantly greater decreases in SLN density than untreated controls (Fig. 9). Other significant differences among herbicides are shown in Table 6.

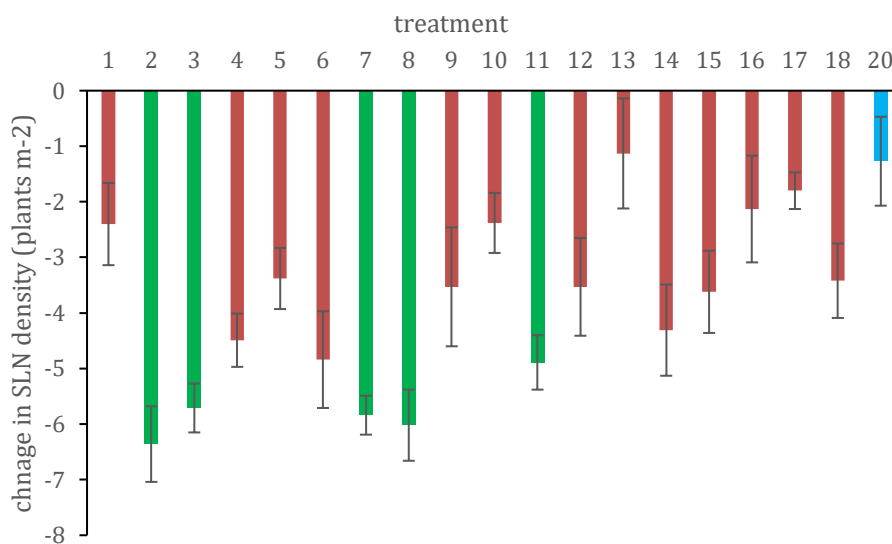


Figure 9. Changes in SLN plant density in the broad-acre trial at Keith in each treatment (monitoring periods combined). Green bars were significantly different from controls (blue bar). ( $p < 0.05$ ). Bars are means  $\pm$  1SE. For values and significant differences among treatments see Table 6.

Table 6. Change in SLN density in each treatment across all monitoring times at the Keith site. Means (SE). Values labelled with different letters were significantly different (Tukey's HSD tests  $P < 0.05$ ).

Treatment	Change in SLN density (plants m <sup>-2</sup> ; (1xSE))	Significance
1	-2.4 (0.74)	BCDEF
2	-6.36 (0.68)	A
3	-5.71 (0.44)	ABC
4	-4.49 (0.48)	ABCDEF
5	-3.38 (0.55)	ABCDEF
6	-4.84 (0.87)	ABCDE
7	-5.84 (0.35)	AB
8	-6.02 (0.64)	A
9	-3.53 (1.07)	ABCDEF
10	-2.38 (0.54)	BCDEF
11	-4.89 (0.49)	ABCD
12	-3.53 (0.88)	ABCDEF
13	-1.13 (0.99)	F
14	-4.31 (0.82)	ABCDEF
15	-3.62 (0.74)	ABCDFE
16	-2.13 (0.96)	CDEF
17	-1.8 (0.33)	DEF
18	-3.42 (0.67)	ABCDEF
20	-1.27 (0.99)	EF

### Warnertown

SLN density was lower at the Warnertown site than at Keith. There were initially  $2.3 \pm 0.2$  (mean, se) plants m<sup>-2</sup>. There was no significant difference in SLN density among plots before treatments were applied (ANOVA  $F = 1.04$ ,  $df = 18$ ,  $P = 0.46$ ). Although the main herbicide effect was not significant (Table 7), there were significant differences in SLN densities among the treatment plots at different monitoring times (time by treatment interaction, Table 8). This interaction shows there were significant differences in the slopes of SLN density versus time over the course of the trial (see Fig. 10). Multiple comparison tests lacked power to detect these differences so changes in SLN density were examined.

Table 7. Results of rmANOVA testing the effects of site and herbicide treatment on SLN density over time in the broad acre trial at Warnertown.

Effects	Value	Num DF	Den DF	Exact F	Pr > F
Treatment	0.01	1	55	0.38	0.54
Time	0.75	3.76	206.8	26.91	< 0.001
Treatment*Time	0.75	3.76	206.8	6.13	< 0.001



## Warnertown

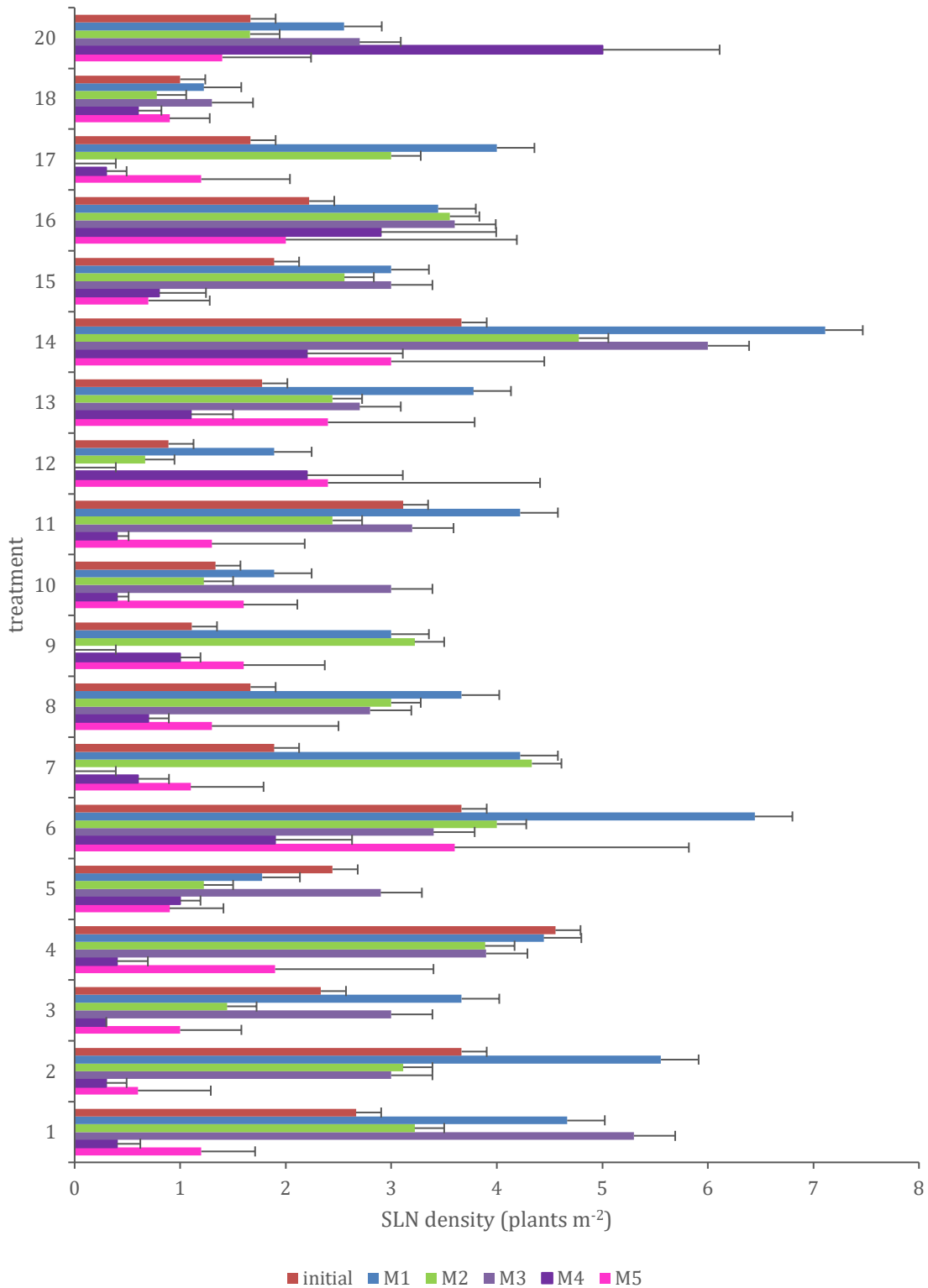


Figure 10. SLN density (mean + 1 SE) in the broad-acre trial before herbicides were applied (initial) and at five successive annual monitoring times at the Warnertown site. Untreated controls are treatment 20. See Table 2 above for identity of other treatments.

The analysis of changes in SLN density, compared to initial values, indicates that there were further significant changes to SLN density across the monitoring period (time by treatment interaction, Table 8).

At Warnertown there was no overall decline in SLN density until the fourth and fifth monitoring times (time effect Table 8, Fig. 11). Treatments 2, 4, 5 and 11 had an overall larger decrease in SLN density over the course of the monitoring period although SLN density was not significantly different to controls (Table 9). Other treatments, including controls had an overall increase in SLN density over the monitoring period (Fig. 12).

Table 8. Results of rmANOVA testing the effects of site and herbicide treatment on changes in initial SLN density over time in the broad acre trial at Warnertown.

Effects	Value	Num DF	Den DF	Exact F	Pr > F
Treatment	0.11	1	55	6.11	0.02
Time	0.54	2.2	119.6	26.75	<0.001
Treatment*Time	0.54	2.2	119.6	4.85	0.008

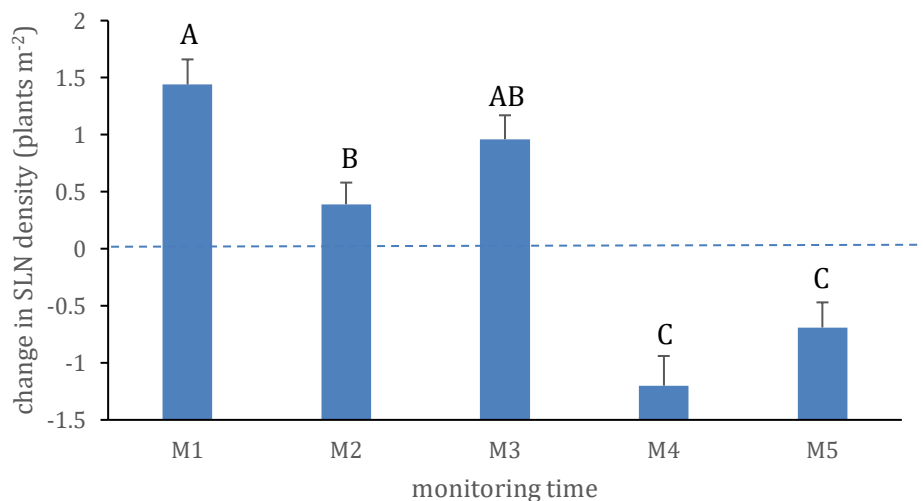


Figure 11. Changes in SLN plant density in the broad-acre trial at Warnertown over the monitoring period (treatments combined). Bars labelled with a different letter were significantly different ( $p < 0.01$ ). Bars are means + 1SE. Values greater than zero indicate an increase in SLN density and negative values a decrease in SLN density.

Table 9. Change in SLN density in each treatment across all monitoring times at the Warnertown site. Means (SE). Values labelled with different letters were significantly different (Tukey's HSD tests  $P < 0.05$ ).

Treatment	Change in SLN density (plants m <sup>-2</sup> ; (1xSE))	Significance
1	0.31 (0.56)	ABC
2	-1.15 (0.75)	AB
3	-0.44 (0.48)	ABC
4	-1.64 (0.5)	A
5	-0.89 (0.39)	ABC
6	0.2 (0.64)	ABC
7	0.82 (0.54)	BC
8	0.62 (0.35)	ABC
9	0.93 (0.34)	C
10	0.29 (0.4)	ABC
11	-0.78 (0.53)	ABC
12	0.93 (0.48)	BC
13	0.71 (0.33)	ABC
14	0.96 (0.59)	BC
15	0.11 (0.35)	ABC
16	0.87 (0.34)	BC
17	0.51 (0.52)	ABC
18	-0.04 (0.21)	ABC
20	0.69 (0.36)	ABC

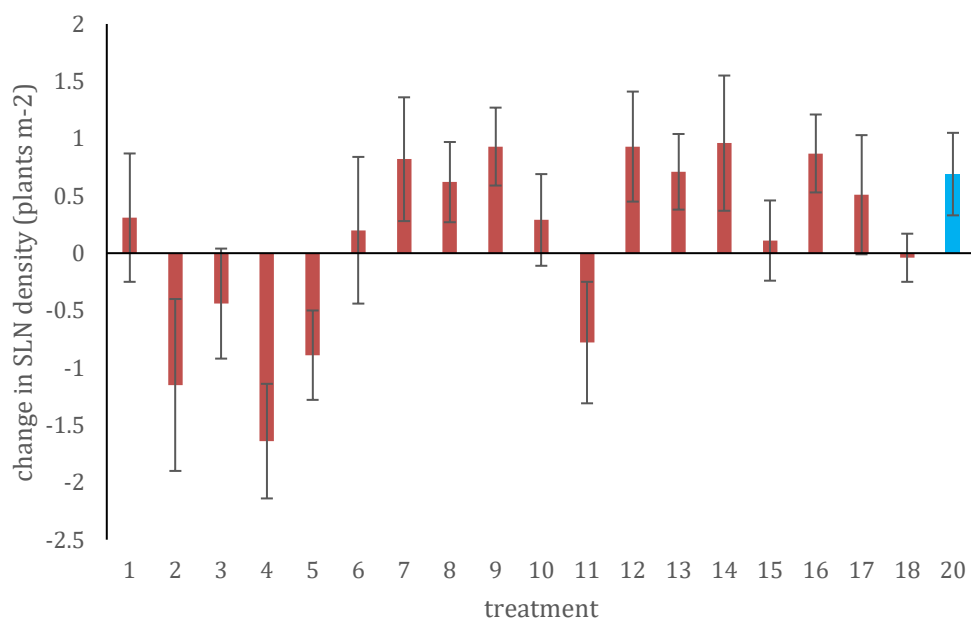


Figure 12. Changes in SLN plant density in the broad-acre trial at Warnertown in each treatment (monitoring periods combined). No treatments were significantly different from controls (blue bar). Bars are means  $\pm$  1SE. For treatment values see Table 9.

## Broad-acre herbicides

The results from both sites illustrate the value of conducting these kind of experiments over an extended period. The pattern of slow revelation of significant results here has been observed in previous research on other perennial weeds. Herbicides were generally less efficacious at Warnertown, probably due to higher water stress on the SLN caused by less September to December rainfall (Fig. 13), warmer temperatures and heavier soils. The beginning of the experiments in early January 2013 unfortunately coincided with a near record heat-wave at both sites (Jan 7 to Jan 13), with daily maximum temperatures between 42 to 46°C. This stressed SLN severely, and resulted in delayed spraying at both sites to allow partial recovery. This situation was repeated in early 2015 when, despite a large rainfall event at both sites in mid-January (Fig. 13), SLN plants were again stressed by at least 5 weeks without significant rainfall in late January and February. It is likely that SLN was under moderate moisture stress at both sites at the time of spraying in early 2014 and early 2015, and this may account for the modest reductions in shoot density measured at both sites. SLN shoots (around two weeks old) were observed at Warnertown on 25 September 2014, demonstrating that SLN perennial shoots can emerge as early as mid-September.

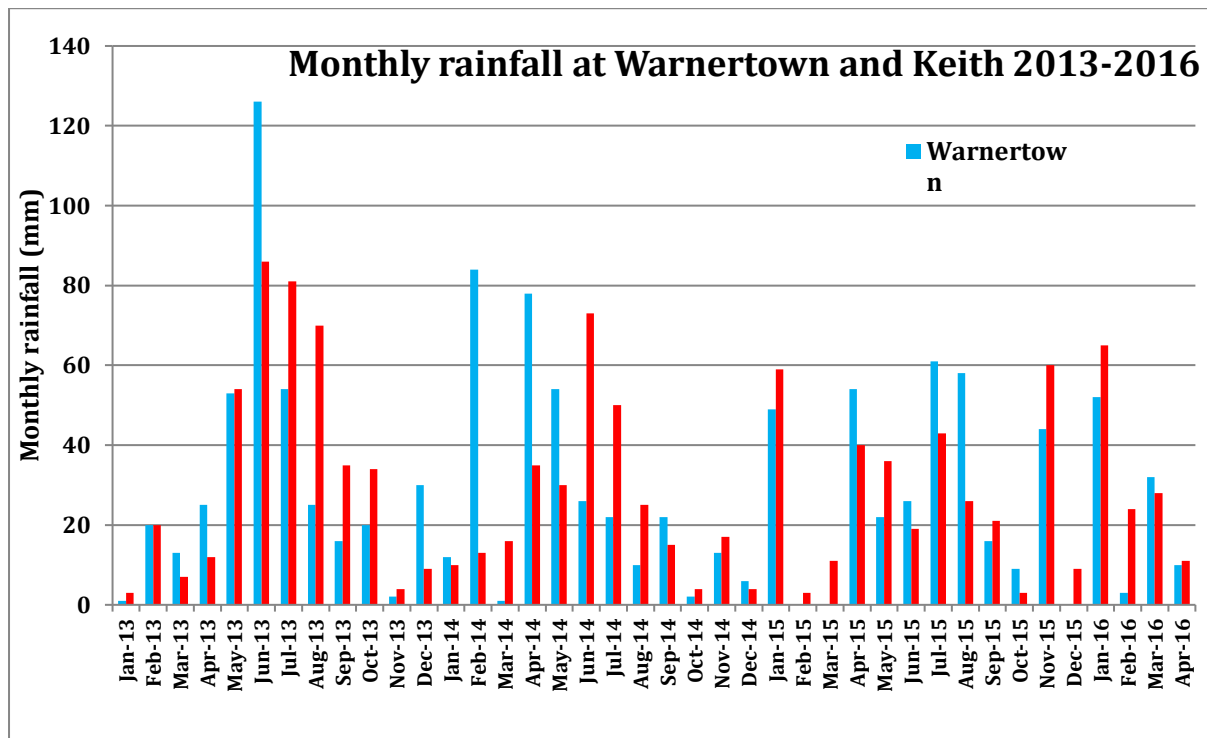


Figure 13. Monthly rainfall (mm) at Warnertown and Keith for 2013 to 2016 spraying.

There were large year to year density fluctuations in untreated control plots at both sites. The patterns were similar at both sites, reflecting general seasonal conditions in South Australia. The fluctuations were probably driven primarily by seasonal changes to available soil moisture, and the patterns seen in untreated plots were reflected in most of the treatments at both sites. Unfortunately these inherent density fluctuations have a confounding effect on statistical analyses. The trial sites chosen had a reasonably even distribution of SLN, and there were no significant differences between plots at the beginning of the experiments at

either site. Keith had a higher initial density ( $8.0 \text{ shoots m}^{-2} \pm 0.3$ ) than Warnertown ( $2.3 \text{ shoots m}^{-2} \pm 0.2$ ).

In summary, the most successful treatments at Keith were treatments 2 (glyphosate  $3 \text{ L ha}^{-1}$ ), 3 (glyphosate  $3 \text{ L ha}^{-1} + \text{EDTA}$ ), 7 (glyphosate  $1.5 \text{ L} + \text{Starane } 0.45 \text{ L ha}^{-1}$ ), 8 (glyphosate  $1.5 \text{ L} + 2,4\text{-D amine } 0.75 \text{ L ha}^{-1}$ ), and 11 (glyphosate  $1.5 \text{ L}$  early then  $3 \text{ L}$  late  $\text{ha}^{-1}$ ). The most successful at Warnertown were treatments 2 (glyphosate  $3 \text{ L ha}^{-1}$ ), 4 (glyphosate  $1.5 \text{ L ha}^{-1}$ ), 5 (Starane  $0.6 \text{ L ha}^{-1}$ ), and 11 (glyphosate  $1.5 \text{ L}$  early then  $3 \text{ L}$  late  $\text{ha}^{-1}$ ). Treatments 2 (glyphosate  $3 \text{ L ha}^{-1}$ ) and 11 (glyphosate  $1.5 \text{ L}$  early then  $3 \text{ L}$  late  $\text{ha}^{-1}$ ) were the only two to perform well at both sites.

The following discussion of specific treatments refers to data in Figures 7, 8 and 9 and Table 6 for the Keith site; and Figures 10, 11, and 12 and Table 9 for the Warnertown site.

**Glyphosate** performed better at Keith than at Warnertown, probably due to higher soil water availability to SLN roots, thus increasing herbicide translocation. Nufarm Glyphosate 540 alone at  $1.5 \text{ L Prod. ha}^{-1}$  was insufficient to control SLN at either site however, with added Pulse penetrant (0.2%), it performed much better and reduced SLN density at both sites, to a level similar to  $3.0 \text{ L Prod. ha}^{-1}$  rate of glyphosate without Pulse. The  $3.0 \text{ L Prod. ha}^{-1}$  rate of glyphosate performed far better than the  $1.5 \text{ L Prod. ha}^{-1}$  rate at both sites, suggesting that the extra investment (c.  $\$8 \text{ ha}^{-1}$ ) is warranted for long-term control. It appears that the likely optimum treatment for SLN would be  $3.0 \text{ L Prod. (540 g/L) ha}^{-1}$  of glyphosate with 0.2 % added Pulse.

The addition of **EDTA** to **glyphosate** was a speculative treatment, based on published reports of high concentrations of  $\text{Ca}^{++}$  ions on the leaf surface of some plant species, and speculation that this might “lock up” some glyphosate before it can be absorbed into the target plant. EDTA is known to bind strongly to  $\text{Ca}^{++}$  ions, and so was chosen in attempt to bind  $\text{Ca}^{++}$  ions on the SLN leaf surface, thus allowing more glyphosate to be absorbed. A concentration of 10 mM EDTA was chosen for the experiment, but Nufarm Glyphosate 540 at  $3.0 \text{ L Prod. ha}^{-1}$  mixed with EDTA performed no better than  $3.0 \text{ L Prod. ha}^{-1}$  alone at either site.

**Starane** (fluroxypyr) at  $600 \text{ ml Prod. ha}^{-1}$  gave a similar level of shoot density reduction to Nufarm Glyphosate 540 at  $1.5 \text{ L Prod. ha}^{-1}$  with added Pulse, at both sites. It performed relatively better at Warnertown than at Keith, suggesting that fluroxypyr may be more robust than glyphosate under dry conditions. Starane at  $600 \text{ ml Prod. ha}^{-1}$  was more effective than Amicide Advance (2,4-D amine) at  $1.5 \text{ L Prod. ha}^{-1}$  at Warnertown, but not Keith. None of these trends were statistically significant. Given that the cost of the two treatments is similar, Starane appears to be a better choice than the long-standing treatment of 2,4-D when SLN is water-stressed. At Keith, where soil moisture is usually higher than at more northerly and westerly regions of SA, glyphosate appears to be more efficacious than Starane, and Starane and 2,4-D gave similar results. This is probably because both glyphosate and 2,4-D are translocated better through SLN roots when soil moisture is higher.

Mixtures of **glyphosate** and **Starane**, and **glyphosate** and **2,4-D amine**, appeared to synergistic at Keith, but antagonistic at Warnertown. At Keith the reduced rates of both Starane and 2,4-D amine, when mixed with Nufarm Glyphosate 540 at  $1.5 \text{ L Prod. ha}^{-1}$ , performed better than label rates of either Starane or 2,4-D amine alone. At Warnertown both mixtures of either Starane or 2,4-D amine with glyphosate gave very poor results. These

treatments should be considered with caution until more data is available, especially when SLN is under moisture stress.

**Hotshot** (aminopyralid/fluroxypyr) gave some suppression at Keith, but failed at Warnertown, and is not considered to be an effective option at this stage.

Nufarm **Glyphosate** 540 at 3.0 L Prod. ha<sup>-1</sup>, followed by **Nuquat** 250 (paraquat) the next day was not as effective as Nufarm Glyphosate 540 at 3.0 L Prod. ha<sup>-1</sup> alone. This was a speculative treatment, designed to try to overcome the problem of herbicide excretion by SLN roots. The treatment explored whether glyphosate might be introduced to the SLN root system by translocation, and then trapped there by the effective “defoliation” of the shoot, using paraquat. The concept is expensive and appears to be unsuccessful, and does not encourage further investigation.

The **split treatments** with an early summer and late summer/autumn component gave no better SLN density reduction than single early treatments alone, at either site. These included Nufarm Glyphosate 540 at 3.0 L Prod. ha<sup>-1</sup> applied in autumn, following summer applications of Nufarm Glyphosate 540 at 1.5 L Prod. ha<sup>-1</sup>, Starane at 450 ml Prod. ha<sup>-1</sup>, or Amicide Advance at 1.0 L Prod. ha<sup>-1</sup>. The combination of dry soil in autumn, and the residual root suppression from the first component of the split treatment, probably restricted the absorption and translocation, and hence efficacy, of the second glyphosate component. These results suggest that the double treatment (“Dual Action”) that is successful in NSW, where soil moisture is higher during summer/autumn, may not be as reliable in SA or WA, or Vic.

**Garlon FallowMaster** (triclopyr) alone at 400 and 800 mL Prod. ha<sup>-1</sup> gave useful suppression at Keith, but failed at Warnertown. It performed better when mixed with glyphosate, resulting in some suppression of shoot density reduction. **Fallowboss Tordon** (picloram/aminopyralid/2,4-D amine) plus additional 2,4-D amine was ineffective.

The treatment comprising Nufarm **Glyphosate** 540 at 1.2 L Prod. ha<sup>-1</sup>, plus **Garlon FallowMaster** at 80 mL Prod. ha<sup>-1</sup>, plus Associate (metsulfuron-methyl) at 7g Prod. ha<sup>-1</sup> was included to represent typical mixtures applied by farmers to control mixed populations of summer weeds (e.g. melons, heliotrope, caltrop etc.). Similar herbicide treatments are incidentally applied to SLN while spraying other summer weeds, with the expectation of at least SLN shoot desiccation. While shoot desiccation is often achieved, the results from both sites suggest that little long-term damage is done to SLN roots.

These experiments have focused on the long-term reduction in SLN shoot density, relying mostly on annual density data from permanent quadrats. Visual effects on SLN shoots several weeks after treatment are variable, and sometimes confounded by drought symptoms. Two reliable sets of data on SLN biomass reduction (visual assessment of % live biomass reduction), 17 and 26 days after herbicide treatments (DAT), were collected at Warnertown in March 2014 and February 2015. These results show that high levels of shoot damage, in the weeks after treatment, do not necessarily correlate with reductions in shoot density. Conditions before and after spraying in 2015 were drier than those in 2014. Glyphosate, in particular, resulted in only slight to moderate visible shoot damage at 17 and 26 DAT. Starane gave excellent shoot control and also reduced shoot density over time. Starane and 2,4-D mixtures with glyphosate also controlled shoot growth quickly. Garlon FallowMaster (triclopyr), Fallowboss Tordon plus additional 2,4-D amine, and the glyphosate Garlon FallowMaster plus Associate mixture all demonstrated effective short term shoot on at least one occasion.

## Spot spray trials

### Keith

The initial density of SLN did not differ among plots (*ANOVA*  $F = 0.35$ ,  $df = 12$ ,  $P = 0.97$ ). There were no SLN plants in plots treated with Uragan or Graslan 8 kg ha<sup>-1</sup> by M3 or Graslan 4kg ha<sup>-1</sup> plots by M5 (Fig. 14). These treatments were excluded from the analysis. In the remaining plots there was a significant difference in SLN density among treatments ( $\chi^2 = 52.3$ ,  $df = 9$ ,  $p < 0.001$ ). The density of SLN plants in several herbicide treatments was significantly less than in control plots in the course of the trial (Table 10).

Arsenal Xpress and FallowBoss Tordon plots had reduced density of SLN at the first monitoring period and density remained low up to four years later (Fig. 14). The lower rate of Hotshot (5 L ha<sup>-1</sup>) was less effective at suppressing SLN than the higher rate (10 L ha<sup>-1</sup>).

Table 10. Parameter estimates from the GLMM comparing model fits of all herbicide treatments with untreated controls (fixed effect) at Keith. Entries in bold indicate the herbicide treatments that differed significantly from untreated controls.

Treatment	Estimate	Std. Error	z value	Pr (> z )
(Intercept)	1.80	0.20941	8.60	< 0.001
<b>Arsenal Xpress</b>	<b>-0.99</b>	<b>0.22943</b>	<b>-4.36</b>	<b>&lt;0.001</b>
<b>FallowBoss</b>	<b>-1.27</b>	<b>0.24346</b>	<b>-5.24</b>	<b>&lt;0.001</b>
Garlon	-0.05	0.19741	-0.29	0.77
Glyphosate	-0.18	0.20054	-0.94	0.35
Glyphosate + EDTA	-0.06	0.19693	-0.31	0.75
<b>Hotshot 10 L</b>	<b>-0.43</b>	<b>0.20727</b>	<b>-2.08</b>	<b>0.04</b>
<b>Hotshot 5 L</b>	<b>-0.41</b>	<b>0.20677</b>	<b>-2.00</b>	<b>0.05</b>
Trimac	-0.05	0.19716	-0.27	0.79
<b>Velmac G</b>	<b>-0.50</b>	<b>0.20918</b>	<b>-2.40</b>	<b>0.02</b>

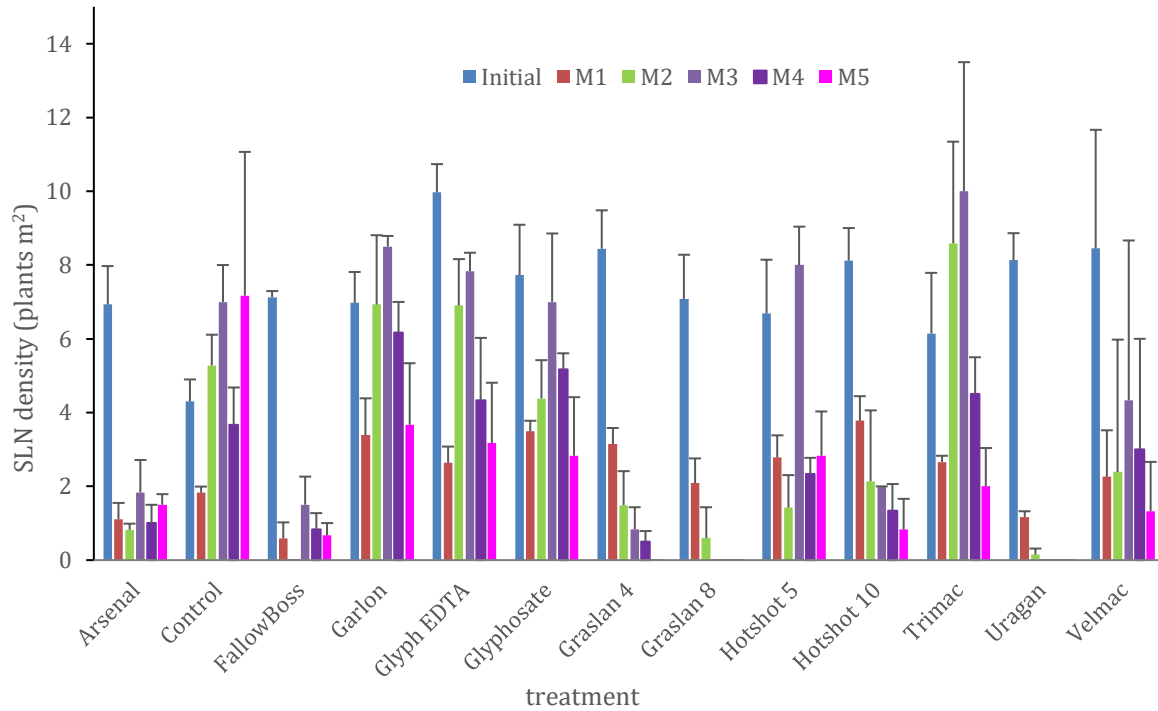


Figure 14. SLN density (mean + 1 SE) before spot spray herbicides were applied (initial) and at five successive annual monitoring times at the Keith site.

#### Warnertown

The initial density of SLN plants did not differ among treatment plots (*ANOVA*  $F = 1.05$ ,  $df = 12$ ,  $P = 0.44$ ). There were no SLN plants in Uragan treatments at the first and subsequent monitoring times (Fig. 15). There were no plants in the Graslan plots ( $8 \text{ kg ha}^{-1}$ ) at or following M2 or in the Graslan  $4 \text{ kg ha}^{-1}$  plots at or following M3. These treatments were not included in the analysis. There were significant differences in SLN density among treatment plots during the course of the trial ( $\chi^2 = 69.5$ ,  $df = 8$ ,  $P < 0.001$ ). All herbicide treatments, with the exception of Trimac plots, had significantly fewer plants than untreated controls (Table 11). These results may be misleading as in several treatments SLN density was very low across all monitoring years. Individual GLMM tests of differences within treatments between years found that only in the Velmac G treatment was there a significant difference in SLN density over time. The low density of plants and variability within plots makes conclusions tentative, but there is some evidence that the herbicide treatments suppressed SLN (Fig. 15). No SLN plants were detected in Uragan treated plots after spraying and plants had not appeared up to four years later. The higher rate of Graslan ( $8 \text{ kg ha}^{-1}$ ) eliminated SLN plants after one year and the lower rate ( $4 \text{ kg ha}^{-1}$ ) after two years, with no plants subsequently occurring in these plots up to four years after spraying. Velmac G reduced SLN density.

Table 11. Parameter estimates from the GLMM comparing model fits of all herbicide treatments with untreated controls (fixed effect) at Warnertown. Entries in bold indicate the herbicide treatments that differed significantly from untreated controls.

	Estimate	Std. Error	z value	Pr ( $> z $ )
(Intercept)	1.49	0.17817	8.37	< 0.001
<b>Arsenal</b>	<b>-1.93</b>	<b>0.34463</b>	<b>-5.60</b>	<b>&lt; 0.001</b>
<b>Garlon</b>	<b>-0.88</b>	<b>0.25441</b>	<b>-3.47</b>	<b>&lt; 0.001</b>



<b>Glyphosate</b>	<b>-1.37</b>	<b>0.28803</b>	<b>-4.74</b>	<b>&lt; 0.001</b>
<b>Glyphostae + EDTA</b>	<b>-1.31</b>	<b>0.28418</b>	<b>-4.61</b>	<b>&lt; 0.001</b>
<b>Hotshot 10 L</b>	<b>-1.11</b>	<b>0.26892</b>	<b>-4.14</b>	<b>&lt; 0.001</b>
<b>Hotshot 5 L</b>	<b>-1.00</b>	<b>0.26192</b>	<b>-3.82</b>	<b>&lt; 0.001</b>
Trimac	0.03	0.21687	0.14	0.89
<b>Velmac G</b>	<b>-1.59</b>	<b>0.30722</b>	<b>-5.17</b>	<b>&lt; 0.001</b>

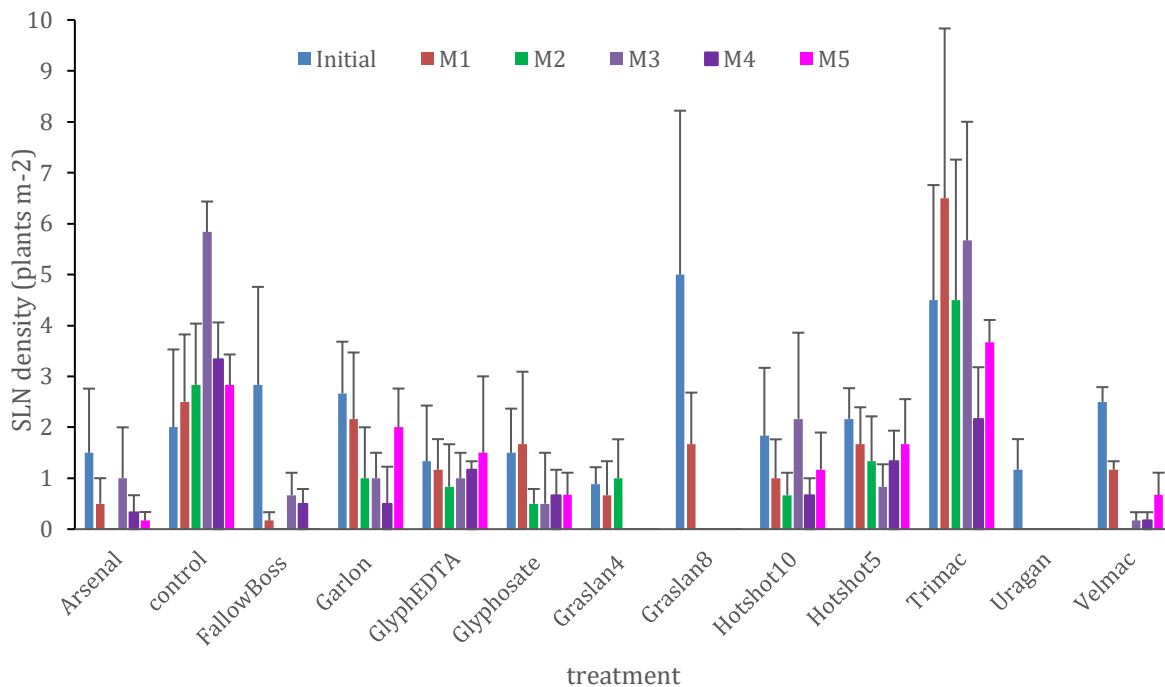


Figure 15. SLN density (mean + 1 SE) before spot spray herbicides were applied (initial) and at five successive annual monitoring times at the Warnertown site.

In summary, Uragan and Graslan were the most effective treatments, and controlled shoot emergence for at least four years after the last treatment. Both are soil residual herbicides that produce bare soil for many years after treatment.

Spot-spraying, or spot-treatment, applications of herbicides are aimed at individual plants or small patches of plants. They are designed to prevent seed set, and to reduce shoot density as quickly as possible. Many of these treatments use herbicides with very persistent soil residues that can produce bare ground for many years. Due to their high cost and residual soil effects on following crops, these treatments are unsuitable for large, established infestations. The largest cost of spot-spraying treatments is often labour, and this increases if several follow-up treatments are required. Consequently, these experiments aimed to treat the experimental plots for two successive seasons, and then measure the effect on SLN density over ensuing seasons. Treatments are shown in Table 3, and reductions in SLN density are shown in Table 10 and Figure 14 for Keith; and Table 11 and Figure 15 for Warnertown.

Meaningful data for spot-spraying treatments needs to be collected over a period of at least five years. The extension of funding for this project has allowed this to happen. While early results can give a good indication of the likely long-term efficacy of treatments, there is no

substitute for field data collected over many years. Treatments generally have one of two major actions – leaf absorption, and translocation through the root system (e.g. glyphosate); or dispersal through the soil, aided by rainfall, and absorption by the roots (e.g. tebuthiuron). The latter action can result in bare ground for a number of years.

As with the results from the broad-acre experiments, the statistically significant results were revealed slowly. Many of the treatments were effective four years after the last of two successive annual treatment applications, demonstrating long-term control. The following discussion of specific treatments refers to data in Table 10 and Figure 14 for Keith; and Table 11 and Figure 15 for Warnertown.

**Uragan** was very effective at both sites, and within one (Warnertown) to three years (Keith) there were no surviving SLN shoots recorded. This treatment appears to have great promise for small patches of SLN, but care should be taken with the effects of prolonged bare soil for many years.

**Graslan** at 4 or 8 kg ha<sup>-1</sup> was also very effective at both sites, but required more time (soil moisture) than Uragan for dispersal through the soil profile. Graslan at 8 kg ha<sup>-1</sup> had completely controlled shoot emergence by two (Warnertown) to three years (Keith), and this was achieved at 4 kg ha<sup>-1</sup> by three (Warnertown) to five years (Keith). Graslan also creates bare soil for prolonged periods.

**FallowBoss Tordon** and **Arsenal Xpress** gave good control at both sites, and may be considered where revegetation of bare soil is required earlier. Although shoots were not completely controlled after two treatments, regrowth over the following seasons has been very well restricted, suggesting good potential for long-term control.

**Hotshot** and **Velmac G** gave useful suppression at both sites, however the level of shoot survival suggests that follow-up applications may be necessary to achieve eradication.

**Glyphosate** gave very useful suppression with limited regrowth at both sites, however the level of control was not as good as with the soil residual treatments such as Uragan and Graslan. However glyphosate treated areas soon revegetate, thus avoiding prolonged bare soil patches.

**Garlon** gave temporary shoot control during the treatment period at Warnertown, however shoot density recovered to an unacceptable level by the end of the monitoring period.

**Trimac** did not give satisfactory control at either site.

## Large plot eradication trials

### Keith

There was a significant difference in SLN density between treatments but not at all monitoring times (treatment X time interaction, Table 12). Initially, before treatments were applied, control and treated plants had the same density of SLN plants (Fig. 16). Density of SLN plants in control plots varied over the five years of the trial but SLN density was consistently lower in treated plots following herbicide application, with the exception of the first monitoring period (Fig. 16).

Table 12. Results of rmANOVA testing the effects of site and herbicide treatment on SLN density over time in the eradication trial at Keith and Warnertown.

Effects	Num DF	Den DF	value	Exact F	Pr > F
Keith					
treatment	1	4	2.68	10.7	0.03
time	1.66	6.65	0.33	7.6	0.02
treatment*time	1.66	6.65	0.33	7.6	0.02
Warnertown					
treatment	1	4	3.09	12.36	0.02
time	1.43	5.71	0.29	7.17	0.03
treatment*time	1.43	5.71	0.29	11.66	0.01

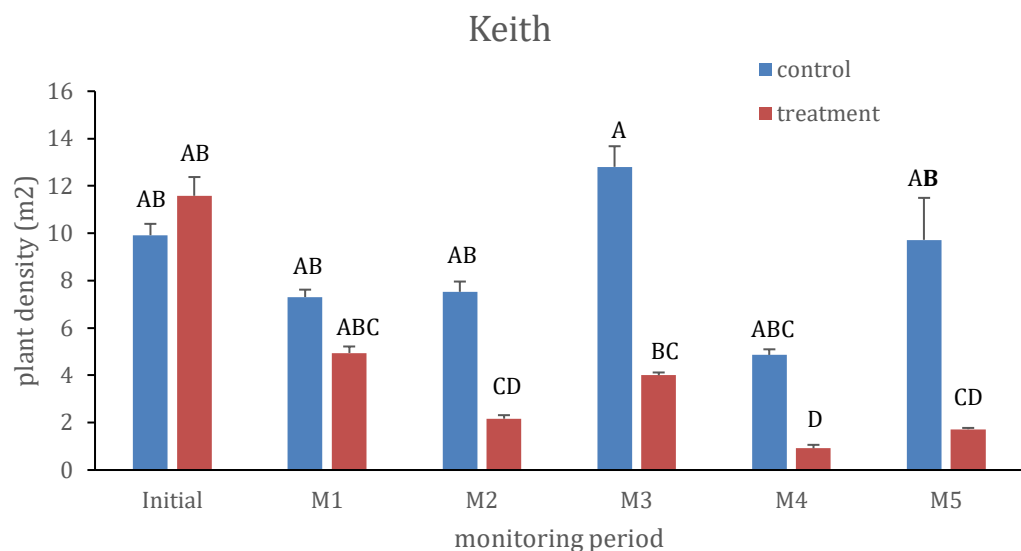


Figure 16. SLN density in eradication plots at Keith in untreated control plots and herbicide treated plots over five successive annual monitoring times. Bars labelled with different letters were significantly different (Tukey HSD test,  $p < 0.05$ ). Bars are means + 1SE.

### Warnertown

At Warnertown there was also a significant difference between treatments but not at all monitoring times (treatment X time interaction, Table 12). SLN density was significantly lower in treated plots than untreated controls at the second and subsequent monitoring times (Fig. 17). SLN density decreased by about 75% after one herbicide treatment and there was a further significant decline by M4.

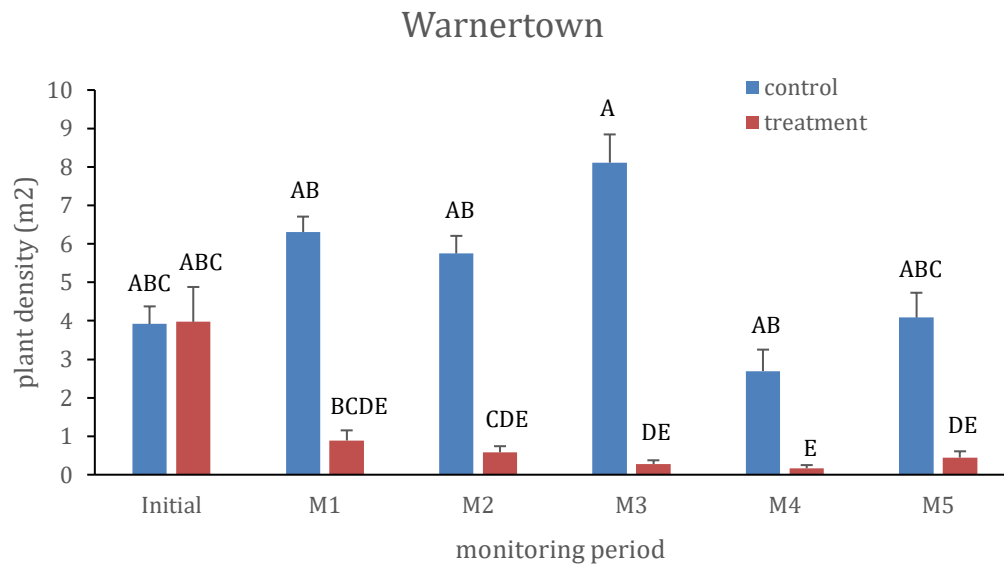


Figure 17. SLN density in eradication plots at Warnertown in untreated control plots and herbicide treated plots over five successive annual monitoring times. Bars labelled with different letters were significantly different (Tukey HSD test,  $p < 0.05$ ). Bars are means + 1SE.

### Stem diameter

The SLN at Warnertown had significantly larger diameter stems than those at Keith (site effect  $F_{1,9} = 5.76$ ,  $p = 0.04$ , Table 13). Stems at Warnertown were an average 4 mm in diameter whereas stems at Keith were an average 3.5 mm diameter. There were significant differences between stem diameters at each monitoring time (time effect  $F_{2,20} = 4.19$ ,  $p = 0.03$ , Table 13). Stem diameters were larger in the year following the first herbicide treatment (mean of 4 mm) but were not significantly different from initial values in the year following the second treatment (mean of 3.5 mm).

Table 13. Results of rmANOVA testing the effects of site and herbicide treatment on SLN stem diameter over time in Warnertown eradication trial.

Effects	Num DF	Den DF	F	Pr > F
Treatment	1	9	0.046	0.835
Site	1	9	5.756	0.040
Time	2	20	4.194	0.030
Treatment*Time	2	20	3.222	0.061

The larger plots (10 x 10m) used in these experiments allowed for more measurements, and hence more precise measurement, of SLN density and stem diameter in response to successive glyphosate treatments. The larger plots were also a good visual demonstration of the results, because the differences are more readily seen than those in 10 x 2m sprayed plots. The results from both sites are very encouraging (Figs. 16 and 17). Three years after the last of three successive annual treatments of 1.62 kg a.i. ha<sup>-1</sup> glyphosate, SLN density has been reduced to low levels at both sites. Three years after the last treatment, SLN density has been reduced by 83% at Keith and by 90% at Warnertown, compared to untreated plots. This demonstrates both that successive applications of glyphosate can significantly reduce SLN density, and that control of shoot emergence is retained for at least three years after the last treatment (Fig. 18).

It should be noted that in an experiment at Edinburgh measuring the effect of dust on leaves on glyphosate efficacy (SAGIT project PIRSA 0113), glyphosate applied at 1.62 kg a.i. ha<sup>-1</sup> reduced SLN shoot density by 88% after only two applications. This rate of decline was faster than expected, and further demonstrates that annual applications of glyphosate at a high rate has great potential to reduce large, established infestations.

Measurement of stem diameter (a proxy for plant size) was undertaken to determine whether the cohort of core surviving shoots were larger than that of untreated population mean. This was to test a theory that small and young shoots were relatively easily controlled and the older, more established SLN shoots were the last to succumb to eradication efforts. Stem diameter remained similar in treated and untreated plots, despite the shoot density declining sharply in treated plots.

SLN density varies naturally between seasons, and it is important to have fixed quadrat positions to account for both spatial and temporal variability. In March 2016, about 11 weeks after the official density measurements had been taken in late December 2015, it was observed that treated plots had a higher density of SLN shoots than untreated in the Keith large plot experiments. The shoots were small, and most had emerged since after the official measurements. During this intervening period two major rainfall events had occurred, totalling 90mm. In this case the new shoots were sprayed during the third annual treatment. This observation has major implications for achieving SLN shoot density reductions, and these and a possible explanation for this phenomenon is explored further in the Discussion section below.



Figure 18. Large plot experiment at Warnertown showing treated (top right) and untreated control plots (top left and bottom left) on January 19 2017, one year after completion of three consecutive annual applications of glyphosate.

## Discussion

### *Broad-acre herbicides*

Farmers typically apply herbicides to large, established infestations of SLN during summer, but SLN is often not the primary target. Summer spraying of paddocks is usually undertaken after significant summer rainfall events, primarily to conserve soil moisture and a manageable seedbed for the following crop. Often summer weeds such as caltrop, heliotrope and melons are the primary targets, and SLN is sprayed incidentally. Herbicide mixtures often reflect these priorities, and costs are reduced by using just enough glyphosate (typically around 650 g a.i. ha<sup>-1</sup>) and partner herbicides to control the annual weeds and to desiccate the SLN shoots. Although application of relatively low rates of glyphosate or Starane may kill above ground SLN shoots, along with annual summer weeds, farmers should be made aware that the additional cost of higher rates (i.e. 1620 g a.i. ha<sup>-1</sup> glyphosate; 600 ml Prod. ha<sup>-1</sup> Starane) may be warranted by the useful reduction in shoot density over several years. Without replicated experimental plots, with untreated controls, this effect will often go unrecognised by farmers.

Glyphosate and 2,4-D amine have been the herbicides most commonly used by farmers in southern Australia to control SLN shoots during summer. These experiments have confirmed

that glyphosate should be applied at a rate of at least 1.62 kg a.i. ha<sup>-1</sup>, and at least once each year. This treatment regime should be expected to reduce SLN density by 80 to 90% after three years. The results have also confirmed the increased efficacy conferred to glyphosate by the addition of Pulse penetrant, and the use of Pulse should be considered as best practice.

Results from this research and NSW research (Hanwen Wu, pers. comm.) has confirmed that Starane (fluroxypyr) should be considered as an alternative to 2,4-D amine. Starane gave excellent shoot control, and useful shoot density reductions over three years, at a competitive cost. It appeared to perform better than 2,4-D at Warnertown, and may be more reliable in drier conditions. Starane has the potential to halt flower and berry development, and to suppress shoot growth, when applied prior to berry formation.

The poor additional control given by a later application of glyphosate at both sites suggests that the “Dual Action” strategy promoted in NSW may not always be successful in South Australia. It is likely that lasting shoot suppression from the early herbicide application (glyphosate, Starane, or 2,4-D amine), added to moisture stress present in many seasons, combine to restrict the absorption and translocation of glyphosate applied in late summer/autumn. The “Dual Action” strategy is more likely succeed in NSW, because summer rainfall and hence soil moisture is often higher.

Throughout these experiments there have been several observed examples of SLN shoot density rising after treatment with herbicides. This is counter-intuitive and needs to be explored. The most likely explanation is that under some circumstances herbicides are translocated some distance into the SLN roots system, killing the lateral root sections closest to the main stem. This may lead to something analogous to “chemical cultivation”, where the damaged plant effectively becomes a collection of disconnected lateral root segments. This can sometimes be observed after mechanical cultivation, where fragmentation and dispersal of root fragments leads to a subsequent increase in shoot density as fragments establish shoots. Adventitious root and shoot buds, previously suppressed by plant growth regulators prior to herbicide application, are released to produce a prolific cohort of new shoots. This phenomenon has important implications for both farmer’s perceptions and long-term SLN control tactics. Firstly, if farmers are unaware of the likely mechanism behind the apparent increase in shoot density, they may conclude that the herbicide has made matters worse and discontinue their use against SLN. More seriously, in some situations herbicides may indeed increase the SLN shoot density if daughter shoots from isolated root fragments are not controlled. This potential situation was observed at Keith in autumn, 2016. Two successive applications of 1620 g.a.i. ha<sup>-1</sup> had led to a rapid decline in shoot density by February 2016, when shoots were counted. However, 90 mm of subsequent rainfall produced a dense cohort of young, small SLN shoots in the treated plots, but not in the untreated. Given the high levels of soil moisture present, it would be very likely that these shoots would develop to become autonomous perennial plants. The implication from this is that beginning and then abandoning long-term management using glyphosate may increase the SLN density, leading to higher yield losses. It will be prudent to warn farmers that once undertaken, it is important to continue until the isolated sections of the root system have exhausted their carbohydrate reserves to the point where adventitious buds can no longer reach the soil surface.

### *Spot-spraying treatments*

Assessment of the efficacy of spot-spraying treatments requires an especially long-term observation time, over many years. Often plants are sprayed once, or perhaps twice, by farmers and then assumed to be “eradicated”. These experimental plots were treated twice, and then shoot density was measured for a further four years.

Uragan (bromacil) and Graslan (tebuthiuron) have emerged as the two most effective spot-treatment herbicides. Both are non-selective pre-emergence soil residual herbicides that are mainly absorbed through the roots, then translocated to the foliage via the xylem to inhibit photosynthesis at the photosystem II pathway. Both also produce bare soil for at least several years when used at high rates. Previous experiments with Graslan have suggested that SLN shoot control lasts for at least eight years, and possibly longer.

Arsenal Express, Uragan, and Velmac G treatments, while relatively effective on SLN, produced totally bare ground. Bare ground can be a problem, because crops are often unable to grow in the residual patches, and livestock tend to “camp” on bare patches, leading to wind erosion and significant hollows where herbicides have been used.

### *Large scale plots*

The rapid decline in SLN density over the first three years was very promising, as was the relative lack of shoot regrowth over the last untreated three years. Once shoot density has been reduced by 80 to 90%, and if cohorts of new shoots arising from disconnected root sections are treated, it may be possible to transition from boom-spraying to spot-spraying.

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# **SAGIT Silver Leaf Nightshade (SLN) Workshop Summary.**

## **Workshops**

Nelshaby Ag Bureaux, 1<sup>st</sup> April 2019

Keith Natural Resources Centre, 9<sup>th</sup> December 2019

## **Attendees**

A total of 35 people attended the two SLN workshops with a mixture of farmers, Natural Resources Management and Local Action Planning staff as well as agronomists in attendance.

The Nelshaby workshop specifically target only landholders with SLN problems although the Keith workshop both farmers and broader land management practitioners.

Landholders in attendance came from Port Pirie, Crystal Brook, Merriton, Gladstone, Warnertown, Keith, Bordertown and surrounds.

## **Content**

A comprehensive presentation was delivered to workshop attendees on the ecology and impacts of silver leaf nightshade (SLN), results of the SLN trials and recommended best practice management approach arising out of the trials. The presentation also included the biological control “journey” and outcome of the project. A range of extension material were provided to participants including the SLN Best Practice Management Manual, SA Weed Control Handbook and other best practice management information.

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In addition to the SLN presentation there was extensive discussion about various farmers experience with SLN and their current approaches, success and failures in managing SLN. Provision of catering provided an opportunity for attendees to not only share experiences regarding SLN management but also discuss management of a variety of other weeds presenting problems to farmers in the Mid-North and South East of the state.

The Keith workshop also provided a presentation on best practice African Lovegrass and African Boxthorn management in addition to demonstration of a “Prickle Picker” and Rotor Wiper which are new pieces of equipment being harnessed for the management of caltrop, African Lovegrass and cape tulip.



**Farmer, agronomists and NRM staff learning about best practice SLN management at Keith**



**Demonstration of new and emerging technologies for the management of declared weeds**

# Invasive Weed Control

## Information Workshop

9 December 2019 12pm – 4pm

Natural Resources South East Office  
61 Anzac Terrace Keith

Workshop/Demonstration on best practice control of

**African Lovegrass**

**Silver Leaf Nightshade**

**African Boxthorn**

Demonstration of a rotor-wiper and prickle-roller

Troy Bowman, Established Weed Facilitator with the Invasive Species Unit (PIRSA) will be presenting on the day, funded through the Agricultural Competitiveness White Paper.



Register by 4th December 2019

BBQ lunch provided.

For more information and to RSVP contact

Tony Richman 0418 893 165 or [tony.richman@sa.gov.au](mailto:tony.richman@sa.gov.au)

[www.naturalresources.sa.gov.au/southeast](http://www.naturalresources.sa.gov.au/southeast)



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