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# **Effectiveness and energy efficiency of pulsed electric microshocks for killing young weeds.**

**A thesis presented in partial fulfilment of the requirements for the degree of**

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**in**

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# Abstract

Alternatives to herbicides are needed due to increased occurrence of herbicide resistance, regulatory restrictions, and consumer preferences. This thesis presents results of research into ultra-low energy weeding systems using very short pulses of very high voltage direct current electricity. Proof-of-concept trials found that small *Capsella bursa-pastoris* weeds in a fallowed vegetable bed were killed by single 10 kV pulses of about 5 J. Grasses were harder to kill. In replicated greenhouse trials, small *Chenopodium album* plants collapsed, and three quarters were dead 4 weeks later. Shocks instantly impacted small *Lolium multiflorum* plants, but death rates were very low.

Applying multiple-pulse treatments using a probe electrode precisely placed against the stems of bagged plants showed *Amaranthus powellii* could be well controlled, but *Solanum nigrum* could generate adventitious roots and recover. While *L. multiflorum* treated with probe-electrodes again had a high survival rate, biomass reduced with increasing energy doses. Seeking a more practical application method, I showed that a flat plate electrode applying multiple pulses to the plants' leaves or to the plant pressed against the soil was effective, with *Polygonum aviculare*, *A. powellii*, *Amaranthus deflexus* and *Solanum nitidibaccatum* weeds successfully controlled. While only half of *L. multiflorum* plants were killed, data indicated a 90% death rate would be achieved at 200 J plant<sup>-1</sup>. Moving outdoors, I treated small plants in the ground using flat plate electrodes and achieved excellent control of *Lepidium didymum*, *A. powellii* and *L. multiflorum*.

Flat-plate electric weeding using pulsed microshocks is a novel development that gives effective control of individual or small clumps of weeds in the field. Observations of plant

responses during almost 30 trials suggest the mode of action is not cell rupturing from resistive heating as usually claimed for electric weeding, but an increase in membrane permeability. My trials showed that small broadleaf weeds and grasses can be controlled at a density of 5 plants  $\text{m}^{-2}$  using less than 1  $\text{MJ ha}^{-1}$ , a fraction of the energy required by any other weeding system. Combined with automation technologies and artificial intelligence, it offers an autonomous, low-energy, non-chemical, selective weeding system for integrated management of weeds.

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## Dedication

I dedicate this work to my whanau; my parents and siblings who instilled belief in and exemplified life-long learning, my partner, children and friends who encouraged and supported my efforts, and to the thousands of researchers who spent tens of thousands of years accumulating the knowledge on which this research is based.

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***Whāia te iti kahurangi, ki te tūohu koe, me he maunga teitei.<sup>1</sup>***

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<sup>1</sup> <https://kupu.maori.nz/kupu/Wh%C4%81ia-te-iti-kahurangi!>



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## List of Abbreviations

A	ampere
AC	alternating current electricity
CO <sub>2</sub>	carbon dioxide
DAT	days after treatment
DC	direct current electricity
EIQ	environmental impact quotient
H <sub>2</sub> O <sub>2</sub>	hydrogen peroxide
ha	hectare
I	current
IWM	integrated weed management
J	joule
kJ	kilojoule
kV	kilovolt
kVA	kilovolt ampere
L	litre
MJ	megajoule
mL	millilitre
ms	millisecond
mV	millivolt
μs	microsecond
ns	nanosecond
nsPEF	nanosecond duration pulsed electric fields
Ω	ohm
PCD	programmed cell death
PEF	pulsed electric field
PMS	pulsed microshocks
ROS	reactive oxygen species
V	volt
W	watt
WAS	weeks after sowing



# Chapter 1. Introduction

## A CHALLENGE PRESENTED

Herbicide resistance management is an increasing issue in New Zealand and globally. It was the topic of a major Ministry for Business Innovation and Employment research programme from 2019 – 2023 (Endeavour Fund C10X1806). Within the overall programme, a review of non-herbicide weed control technologies was proposed, and that was the genesis of this PhD research journey. Three technologies were proposed as candidates for assessment: air-blast abrasion, hot-water/hot-foam, and electrocution. The context for technology application was managing herbicide resistant weeds in vegetable and arable crops, notably controlling “escape weeds” following a herbicide or mechanical weeding operation.

## 1.1 Technology selection

A loosely defined selection matrix was used to compare and select non-herbicide weed control technologies for further investigation and development.

With carbon accounting increasingly part of business (Danley, 2023) and spikes in energy costs (Ministry for Business Innovation and Employment, 2019), energy requirement was a key factor for consideration, and the acknowledgement that minimal tillage and regenerative farming practices typically result in higher surface residue levels showed a system needed to cope in such a situation. Labour shortages and costs coupled with rapid advances in farm automation pointed to a system that could be deployed robotically with minimal supervision.

The final unweighted selection list included consideration of:

- Applicability to vegetable and arable cropping
- Ability to control herbicide resistant escape weeds surviving a prior treatment
- Effectiveness and energy efficiency in a field situation
- Ability to be used selectively
- Ability to cope with surface residues
- Minimal requirement for consumptive inputs
- Suitability for robotic deployment
- Practically: equipment had to be available for testing.

The three nominated technologies were reviewed within this selection frame, and assessments are summarised below.

### **1.1.1 Air-blast abrasion**

Abrasion equipment for research was built by AgResearch to a USA design (Figure 1-1).



Figure 1-1 Photo of the handcart-mounted abrasion weeding equipment provided by AgResearch, showing the mobile compressor (top left) and compressed air tank (centre), grit hopper and feed mechanism (right) and delivery lance (foreground).

A number of abrasive materials were tested including ground dried sheep manure, sawdust, pumice sand, ground macadamia shell, fine garnet, paving sand, and ground oyster shell. The same type of equipment has a long history of success in USA research (Forcella, 2009a, 2017; Forcella et al., 2023) using a range of products, including walnut, corn grits and other readily available farm waste materials.

However, satisfactory weed control was not achieved using the provided equipment or the abrasives tested. The most effective abrasives were garnet and paving sand which are increasingly recognised as limited resources and were only able to damage leaf tissue, leaving bruising and pockmarks, but seldom removing leaves or stems with growing points. Close up photographs of some blasted leaves are shown in Figure 1-2.



Figure 1-2 Photographs of leaves subjected to blasts from the abrasion weeder showing pockmarked and bruised leaves (A) *Taraxacum officinale* (dandelion) and (B) *Plantago lanceolata* (narrow leaved plantain) and *Lolium perenne* (perennial ryegrass).

The failure to demonstrate effective weed control in preliminary testing, combined with the requirement for finite consumables, high CO<sub>2</sub> emissions from the high energy demand to compress air and the production and transport of abrasives (Corrosion Alliance, 2024), led to the technology being dismissed from further research.

### 1.1.2 Hot foam

A commercial Foamstream M600 hot-foam system (Foamstream Ltd., Co. Laois, Ireland) was located, and a demonstration arranged (Figure 1-3).



Figure 1-3 Photograph of a hot foam system being demonstrated at the Centre for Land and Water in Hastings, NZ showing the foaming application nozzle and the utility-mounted diesel-powered boiler and water tank. (Trevor James image)

Martelloni et al. (2019) presented an image of a very similar machine and of a foam strip laid across the ground, and described research showing that doses of between 33,000 and 80,000 L ha<sup>-1</sup> were required. Hatcher and Froud-Williams (2017) also presented images that showed a large volume of water with patchy foam coverage. Together, these images suggested that environmental conditions, operator training and experience could influence the effectiveness and efficiency of this technology. Poor weed control, the need to transport very large volumes of water around paddocks, an extremely high diesel-energy demand, assuming a 65% aqueous air solution (Tindall et al., 2009), and no obvious potential for selectively treating individual plants (targeted weeds) led to this option being dismissed.

### 1.1.3 High-energy electric weeding

Initial scoping found only one electric weeding system available in New Zealand. This was a Rootwave Pro™ spot-weeder and demonstrations by HotGrass Ltd. were arranged for field day events in Hastings and in Pukekohe (Figure 1-4).

The Rootwave Pro™ system has a relatively high energy demand, requiring a 230 V single-phase generator with 7.5-10 kVA capacity. It was relatively expensive to buy and operate and, while of potential use in an urban environment with ability to electrocute large weeds, it showed minimal suitability as a broadacre weed management technology for controlling escape weeds. It was tested by Christchurch City Council, but was not selected for ongoing use.



Figure 1-4 Photograph of the Rootwave Pro™ system by HotGrass Ltd. At a Pukekohe, NZ demonstration of showing the treatment electrode, heavy duty electric cable, trailer with generator, and operator wearing electrically tested insulating gloves and footwear.

There are many other electric weeding machines available, with first patents in the 1890s (Sharp, 1893; Scheible, 1895). Patents have continued to be granted with notable interest in the 1970s and 1980s (Dykes, 1977; Tibbs, 1977; Gilmore, 1981; Laronze, 1981), and continuing through to the present (Diprose, 2016; Pomilio et al., 2019; Claver, 2023). Farmers continue to use Lasco™ equipment developed in the 1970s (Bennett, 2019) and Ubiquitek has a range of new machines (Claver, 2023; Cordis, 2024) building on the early work of Diprose (2016). Slaven et al. (2023) are testing the Zasso™ XPower machine in Western Australia, and Rowland et al. (2023) are testing the 6R30 Weed Zapper™ at Cornell University in Ithaca, NY. Notably, these are expensive, high energy machines running large generators and best suited to broadacre or strip weeding applications. Because of the high cost and energy demand, limited ability for selective weed control in an automated system, and no equipment being available to test, the high-energy electrical systems were also dismissed.

#### **1.1.4 Low-energy electric weeding**

Literature reviews found references to very low energy electric weeding, initially the Japanese research and patents by Mizuno and colleagues (Mizuno & Hori, 1988; Mizuno et al., 1990; Kimura & Mizuno, 1992; Mizuno et al., 1993; Ejima et al., 1997). These reported successful weed destruction using orders-of-magnitude less energy than other electrical, or indeed any other, weeding systems (see Chapter 2 Table 2-1 from Bloomer et al. (2023b)). Further searching found references to Russian research into low-energy electric weeding that had been continuing since the 1970s (Baev & Savchuck, 1974; Baev & Yudaev, 2017) and to very low-energy weeding research by Blasco et al. (2002). Recently, Abdelghafour et al. (2021); Lati et al. (2021) and Lehnhoff et al. (2022) also published complementary research using low energy electricity.

### **1.1.5 Potential for robotic deployment**

The potential for low-energy electric weeding technology to be deployed robotically was demonstrated on a study trip I made to Europe in 2016. I visited Ecorobotix, a startup agritech company in Switzerland, which was at that time developing its first generation solar powered weeding robot (Vaqar, 2018; Agtecher, 2024). The Generation 1 Ecorobotix system used plant recognition algorithms to discriminate between crop plants and other plants (weeds). Mechanical cultivators and spot-spray nozzles were being trialled, but the machine appeared suitable for a low-energy electric application system using precisely placed electrodes, which would fit the low-powered solar robot. When I began this PhD research, the Ecorobotix style of robot appeared suitable for very high efficiency electric spot weeding, such as for escape weeds following treatment with a herbicide, so long as a suitable treatment electrode and actuation method were available. Abdelghafour et al. (2021) presented research using the Ecorobotix machine with an electric probe device. They used a thin-rod-probe at 40 – 60 kV AC with 0.25 – 1.0 s pulses and reported only 13 – 75% effectiveness in greenhouse trials depending on species, because they struggled to achieve the necessary precision of probe placement in the field. A prototype robot using electric weeding was developed by Dang (2009), with a 22 kV treatment applied by a suspended electrode. It did not appear to progress beyond a research project. In recent months, several other researchers have referenced my published papers presented here as Chapters 2 – 5 (Bloomer et al., 2022, 2023a, 2023b, 2024). Notably, the “Robots and Shocks” paper (Chapter 2) is cited by Nguyen and Tung (2024) who present details plans for “building a DELTA robot that is specifically engineered to eliminate weeds in agricultural environments”. The system they describe is virtually identical to the Generation 1 Ecorobotix machine.

## 1.2 Research Aims

Having assessed and dismissed abrasion, hot-foam, and high-energy electrical weeding methods, and considering the energy demands and suitability for selectively targeting escape weeds in vegetable and arable crops with significant quantities of surface residues, my focus became high-efficiency, high-voltage electric-weeding systems. As described in Chapter 3:

*The focus of my research is efficient weed control with minimum energy requirements achievable by application of precisely applied microshocks with precisely controlled direct current (DC) voltage, pulse number, pulse length and period (hereafter PMS) to deliver low energy electric treatments. This would make a simple battery-powered hand-held weeding system or high-efficiency strip-weeders possible. In conjunction with the other factors, the technology would be suitable for deployment on low-energy autonomous field robots. It would address issues of herbicide resistance, consumer desire for spray-free food, and avoid chemical residues in water.*

## 1.3 Research objectives

To guide this research, three key objectives were established:

1. Define a set of performance criteria for a novel weeding device in response to increasing herbicide resistance, consumer desire for spray-free food, and a move towards regenerative agriculture in crop production.
2. Determine the minimum dose required to kill a range of small broadleaf and ryegrass weeds in a laboratory greenhouse setting.
3. Identify a practical application solution that could be employed to selectively apply such doses to small broadleaf and ryegrass weeds in the field.

Having shown that PMS weeding was effective with only a fraction of the energy needed by other systems, a fourth objective was added:

4. Review the literature on possible electric weeding modes of action and propose mechanisms that may enable the greatly reduced energy requirements of pulsed multishock treatments

## **1.4 Research goals**

Seeking a measure by which success might be assessed, a goal was set of successfully killing more than 90% of targeted weeds using less than 2 MJ ha<sup>-1</sup> application energy at 5 plants m<sup>-2</sup>, using a system able to selectively target weeds in vegetable and arable crops with significant surface residues.

Is 90% control an adequate target? Clearly 100% control is better, as the more weeds that survive any treatment, the greater the possibility of population increase and of a resistance issue emerging. The 90% target was considered reasonable based on the work of Nørremark et al. (2006) who reviewed reported levels of dry matter reduction of annual and perennial monocot and dicot weeds subjected to physical damage in pot experiments.

Application energy of < 2 MJ ha<sup>-1</sup> is 10% of the energy target suggested by Coleman et al. (2019) for post-weeding escapes, and 5% of the target set by Nørremark et al. (2006) for autonomous inter-row weeding. Note however, that those authors include the total energy need including that for transporting equipment around a paddock, activating any actuator equipment such as robotic arms, and any navigation, vision and computing power required for successful operation.

The nominated 5 plants m<sup>-2</sup> weed density is considered reasonable for post weeding escapes after treatment by other means ((Kaufman & Schaffner, 1982; Vigneault et al., 1990; Coleman et al., 2019).

## 1.5 Proof of concept

A serendipitous event linked my research with a small group of students who were designing prototype electric weeding equipment as part of an engineering project at Massey University (Harvey et al., 2019). On his graduation, one of the students, Hamish Penny, was commissioned to develop ultra-low energy weeding equipment. He established a company (WedaTech Ltd, Hastings, NZ), designed, and built the direct current (DC) systems used in this research. These devices and their preliminary testing are presented in Chapter 3. The first WedaTech system was a single pulse, “spark” weeder operating at about 3, 6 or 10 kV (Figure 1-5).



Figure 1-5 Photograph of Hamish Penny holding a WedaTech Zapper V1.0, a highly mobile weeder running off a small lithium battery and able to apply single-pulse DC pulses of about 5 J. The aluminium-rod treatment electrode is at the end of the black arm and the aluminium earthing electrode is inserted into the soil and grounded via the white wire.

The spark weeder was used in preliminary trials with promising results (Chapter 3 and Chapter 6), including application of shocks to flatweeds germinating in a fallowed vegetable bed (see Figure 6-3 to Figure 6-5). Treated species included *Capsella bursa-pastoris* L. (shepherd's purse), *Sonchus oleraceus* L. (sow thistle), *Lepidium didymum* L. (twin cress) and *Poa annua* L. (annual poa). Treatments of 5 J killed small weeds and 15 J killed most treated plants.

Early trials (described in Chapter 3) applying various “doses” of energy to bag-grown plants in a greenhouse showed the strong single pulse of the spark weeder was of limited effect on small ryegrass weeds, because the fine leaves disintegrated in a manner likened to an electric fuse blowing. This seemed to protect the main stem and growing points and although various deformation symptoms appeared, including leaves with a concertina folding (Figure 3-3) or with tips trapped in the sheath while the blade continued to elongate (Figure 3-4), treated plants often survived. Therefore, a second system was developed, which was able to apply multiple, extremely low-energy pulses with the discharge voltage, pulse length, duration and period able to be controlled by a connected laptop computer (Figure 3-1). The system also included a Pico® high-voltage differential probe and Pico® oscilloscope to measure and record each pulse, allowing detailed electrical data to be collected automatically (Figure 3-2). The multiple pulse approach did not altogether stop the leaf “blowing the fuse” when using probe electrodes at higher voltages, but in greenhouse trials it generally enabled better weed control at lower 3.0 – 3.5 kV treatments.

## 1.6 Thesis Structure

This thesis is in seven chapters. Following this introductory chapter are four chapters from papers published in peer reviewed journals of good standing, an unpublished chapter with discussion of observations and possible modes of action, and a final chapter of concluding discussion that draws the salient points and key findings together.

Chapter 2: *Building a Case* was published in the *New Zealand Journal of Agricultural Research* as “Robots and shocks: Emerging non-herbicide weed control options for vegetable and arable cropping” (Bloomer et al., 2023b). It is a literature review that contextualises non-herbicide weed management for New Zealand farming in the 21<sup>st</sup> century. While herbicide resistance is undoubtedly a driver, consumer preference for spray-free food, increasing demands for regeneratively grown, safe, high-quality produce, awareness of environmental impacts, and local and global regulation increasingly restricting agrichemical use are also forcing changes to weed management strategies. But the emergence of agritechnologies incorporating automation, machine vision and artificial intelligence, and development of new techniques for weed destruction, offer alternatives that minimise or avoid the requirement for herbicides, avoid soil disturbance and can work effectively in high crop or crop-residue conditions.

Chapter 3 *Proof of Concept* was published in *Agronomy* as “Micro electric shocks control broadleaved and grass weeds” (Bloomer et al., 2022). The chapter describes proof-of-concept research using the novel electric-weeding technology developed for this project. Greenhouse trials used two pulse generation systems, one single and one multiple-pulse, and demonstrated that the application of precisely applied microshocks with precisely controlled direct current (DC) voltage, pulse number, pulse length and period can kill

small *Lolium multiflorum* Lam., *Chenopodium album* L., *Amaranthus powellii* S. Wats. and *Solanum nigrum* L. plants using minimal energy.

Chapter 4 *Practical Application* was published in *Agronomy* as “The Electric Spatula: Killing weeds with pulsed microshocks from a flat-plate electrode” (Bloomer et al., 2023a). The Chapter describes further research which tested a system that could be deployed via a hand-held unit or as part of a fully automated system to control escape weeds in field crops. This research showed that *Polygonum aviculare* L. (wireweed), *A. powellii*, *Amaranthus deflexus* L. (prostrate amaranth) and *Solanum nitidibaccatum* Bitter (hairy nightshade) plants were successfully controlled, with the energy required to kill 100% of seedlings varying from 0.1 to 0.9 MJ ha<sup>-1</sup> and indicated that *L. multiflorum* could also be effectively controlled by a treatment of < 1.0 MJ ha<sup>-1</sup>.

Chapter 5 *Field Deployment* was published in *Sustainability* as “Pots to plots: Microshock weed control is an effective and energy efficient option in the field” (Bloomer et al., 2024). This progressed the research from the greenhouse (laboratory) to the farm, testing the weed killing effectiveness and energy efficiency of a flat-plate electrode applying pulsed direct-current electric microshocks (PMS) to plants in bags and in the field. Applying the greenhouse-proven system in the soil outdoors was as effective and efficient as in-bag, greenhouse trials. Better than 90% control was achieved for both *L. didymum* and *A. powellii* at 15 kJ ha<sup>-1</sup>, and for in-ground *L. multiflorum* at 555 kJ ha<sup>-1</sup>, all well below a revised target of 1 MJ ha<sup>-1</sup>. Additional investigation considered the effect of earthing electrode types and of increasing earthing and treatment electrode separation distances. A thin dry-soil layer was found to increase efficiency of soil-pressed flat-plate weeding by reducing energy losses direct to soil.

Chapter 6 *Modes of action* has not been published. It presents observations of plants' immediate responses to applied treatments and their ongoing appearance for days and weeks afterwards and considers how the effects of high intensity pulsed electric field treatments on weeds can be so much more energy efficient than other methods. The chapter is descriptive and pictorial, comparing observations made over almost 30 trials with plant science, electric weeding, electro-medical, and food processing literature. Possible cellular and tissue responses to PMS applied to weed seedlings are proposed. Non-thermal effects, including membrane electroporation, vascular damage and programmed cell death are suggested as potential biological processes involved in plant death, or in cases where treatments were non-lethal, in plant recovery.

Chapter 7 *Concluding discussion* presents lessons and directions from 5 years of ultra-low energy electric weeding research, drawing the material from almost 30 trials together. Research objectives and progression through a series of trials are summarised, and research outcomes describing the fundamental new knowledge gained are presented. Finally, opportunities for further research and development are identified and a path to commercial deployment considered.

# Chapter 2. Building a Case

LOW-ENERGY ELECTRIC WEEDING IS AN OPTION FOR NON-HERBICIDE WEED CONTROL IN VEGETABLE AND ARABLE CROPS

## 2.1 Publication

This chapter was published as:

Bloomer, D. J., Harrington, K. C., Ghanizadeh, H., & James, T. K. (2023). Robots and shocks: emerging non-herbicide weed control options for vegetable and arable cropping. *New Zealand Journal of Agricultural Research*, 67(1), 81-103. <https://doi.org/10.1080/00288233.2023.2252769> (Bloomer et al., 2023b).

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This paper uniquely brought together literature reviews of consumer expectations of vegetable and arable crop farming in New Zealand, the regulatory and market pressures for change, increased evolution of herbicide resistance, and advances in farm equipment automation and technologies for controlling weeds without herbicides. It proposed a future approach to weed management and set a platform for further research and development of automated ultralow-energy electric weeding within an integrated weed management system.

**Statements of Contribution:** See Appendix 1 Statements of Contribution

## 2.2 Abstract

For decades, herbicides have provided easy-to-use, cost-effective weed management, but alternatives are desired. Consumer preference for chemical-free food, awareness of environmental impacts, regulation increasingly restricting agrichemical use, and increasing prevalence of herbicide resistance are forcing changes to weed management strategies. New Zealand farming must remain sustainable and profitable while responding to changes in its overseas markets, among which demands are increasing for regeneratively grown, safe, high-quality produce. Current reliance on herbicides should be reduced, with more emphasis on preventative management by cultural means, and weed suppression by alternative technologies. The emergence of agritechnologies incorporating automation, machine vision and artificial intelligence, and development of new techniques for weed destruction, offer alternatives that minimise or avoid the requirement for herbicides, avoid soil disturbance and can work effectively in high crop or crop-residue conditions. I have identified electric weeding as a feasible alternative and pulsed electric microshocks as a very low-energy option requiring a fraction of the energy of any other system. Pulsed microshocks enable an integrated weed management system for vegetable and arable crop production that combines cultural controls and inexpensive pre-planting treatments with automated application of chemical-free in-crop weed control. Open-source software enables community development of autonomous deployment for niche crops.

**Keywords:** weed control; integrated weed management; regenerative agriculture; alternative weeding; automation; electric weeding; machine vision; artificial intelligence.

## 2.3 Introduction

Weed control is essential for reliable yields of quality produce, as weeds are the major cause of yield loss in key crops such as maize, wheat, potato, rice, soybean (Oerke, 2006), and maize (James et al., 2000). They impact product quality and make farm operations more difficult (McErlich & Boydston, 2014). For decades, chemical herbicides have provided cost-effective weed management. Their ease of use has enabled chemical technology to become the dominant weed control technique, with an array of choices for broad-spectrum or selective control of weeds, even from within standing crops. Weeds of any size can be killed, from newly germinating seedlings to large trees. Intra-row weeds can be effectively removed with minimal if any crop damage. However, the dominance of herbicides is challenged.

A desire for healthier food, soil, and ecosystems, consumer preference to avoid the use of chemicals, and increasing prevalence of herbicide resistance, are driving efforts to find alternatives. At the same time, social challenges to current production systems stemming from an awareness of carbon-balance impacts to climate change, and increased interest in conservation and regenerative agriculture are compounded by labour shortages and increased energy costs. The cost of weed control in New Zealand seed and grain crops - arable including maize - was estimated in 2017 to be c. \$18.4 million per annum or about \$94 ha<sup>-1</sup> (Saunders et al., 2017). For specialty and vegetable crops the costs can be even higher, especially if hand-labour is required (Fennimore & Cutulle, 2019). Political and market requirements impacting weed control include more stringent regulation, restrictions on the use of herbicides, restrictions on burning crop residues, the introduction of carbon accounting, and the promotion of alternative farming systems including regenerative agriculture (Ministry for Primary Industries,

2022). An integrated weed management system must evolve that recognises such requirements and suits production from farms that place more emphasis on regenerative practices, energy efficiency, reduced use of chemicals and maintenance of soil cover.

Recent years have seen many weed control approaches revisited. As a result of research, integrated weed management is recommended, although there is a suggestion that on-farm uptake of this is low (Moss, 2008). Accelerating development of automation and vision systems coupled with artificial intelligence is allowing old and new weed control technologies to be used in hitherto impossible ways. In 2019, we reviewed and conducted preliminary tests (unpublished) with a commercial hot foam/hot water weeding system, and a custom-manufactured air-blast grit system based on the work of Forcella (2009a, 2017). The hot foam/hot water system was rejected on the basis of the extremely large volume of water requiring heating and transport, the energy required (De Cauwer et al., 2015; De Cauwer et al., 2016), the impracticality of the system, as demonstrated, to be effectively deployed in a cropping system, and its inability to achieve selective weeding. The abrasion system provided was tested using a range of grit types, including macadamia shell, oyster shell, coffee grounds and fine sand. None proved totally effective. Fine sharp sand caused most damage, but generally just pock-marked the treated leaf tissue rather than destroying it. Our experience contrasted to the US research that has demonstrated good control in many trials over many years (Forcella, 2009b; Forcella et al., 2011; Forcella, 2019). However, the approach was discounted as it requires heavy equipment and materials, and air compression demands high energy.

Electrical weeding technologies were also investigated. Literature searches identified work demonstrating that very low energy electrical weeding is possible (Mizuno & Hori, 1988; Mizuno et al., 1990; Blasco et al., 2002) and a chance meeting

with a group of engineering students at my university offered the possibility to develop and test such a system. My first results showed the system can be successful in greenhouse trials (Bloomer et al., 2022) and indicated that field deployment is viable as part of a manual or robotic system.

This chapter reviews vegetable and arable crop weed management in a New Zealand context, reviews the range of new and not-so-new technologies available and considers how weed management can adapt to the new context in which vegetable and arable production operates. One concept, pulsed electric microshocks (PMS), is an example of a novel approach for selective weeding within a multimethod, integrated weed management system. This ultralow-energy weeding method can be integrated with automation technologies and artificial intelligence to create an autonomous ultralow-energy, selective, non-chemical weeding system.

## **2.4 New Zealand farming environment in 2023**

Consumer preferences for organic or chemical-free food (Magnusson et al., 2003; Devcich et al., 2007; Forbes et al., 2009; Rutledge, 2009; Crinnion, 2010; Wooliscroft et al., 2014; Hoek et al., 2017; Koch et al., 2017; Galati et al., 2019), public concern about residues in water (Bajwa et al., 2015; Hageman et al., 2019) including public perceptions in Twitter “heavily skewed toward negative sentiments” (Jun et al., 2023), awareness of potential impacts of chemical treatments on soil microflora (Helander et al., 2018) and fauna (Salminen et al., 1996) and restrictions or bans on products including glyphosate (Beckie et al., 2020; Alcántara-de la Cruz et al., 2021), further promote herbicide alternatives. Labour shortages (Kitchin, 2021; Giovannetti, 2022; Ministry for Business Innovation and Employment, 2023; Statistics New Zealand, 2023) force a reduction in manual weed management and drive demand for technology

to replace it. Spikes in the costs of diesel, electricity, fertilisers, chemicals and compliance are forcing farmers to ensure all costs are tightly managed (Murphy, 2022).

Regenerative agriculture, which places emphasis on sequestering carbon and reducing the use of chemicals (Grelet & Lang, 2021; Ministry for Primary Industries, 2022; Schlesinger, 2022; Thomas, 2022; Danley, 2023; McCain Foods, 2023), has re-emerged as a trend supported by governments and industry (Thomas, 2022). In New Zealand, it is seen as a production system focused on reducing the impacts of food production on the environment, a way to shift to low-emissions and a sustainable economy, and is part of the Government's and primary sectors' "Fit for a Better World" roadmap (Ministry for Primary Industries, 2022). A bundle of principles rather than a set of prescribed rules, and variously described (Grelet & Lang, 2021; Ministry for Primary Industries, 2022; Thomas, 2022; McCain Foods, 2023), regenerative agriculture advocates minimising soil disturbance, keeping living roots in the soil, enhancing biodiversity, reducing the use and impact of agrochemicals and artificial fertilisers, and integrating animals into the system. Regenerative agriculture seeks to keep the soil covered with living crops or crop residues covering the soil as much as possible (Thomas, 2022; McCain Foods, 2023). However, in a study of USA corn production, LaCanne and Lundgren (2018) found regenerative agriculture systems had 29% lower grain production but 78% higher profitability than conventional production systems. The movement towards conservation agriculture, systems that are typically reliant on herbicides (Melander et al., 2013; Reiser et al., 2019), is also accompanied by greater quantities of ground cover in the form of crops, cover crops or crop residues, and possibly living mulches within main crops (Westbrook et al., 2022). This makes mechanical weed control by tillage difficult, so non-contact methods are preferable (Bauer et al., 2020) and equipment used should operate effectively in high residue

conditions.

Herbicide resistance, “the inherited ability of an individual plant to survive a herbicide application that would kill a normal population of the same species” (Peltzer, 2019), is increasing globally (Wilson et al., 2011; Owen et al., 2014; Powles, 2014; Walsh & Powles, 2014; Lamichhane et al., 2017; Moss, 2017; Heap, 2020). Herbicide resistance has been increasingly reported in New Zealand (Rahman et al., 1983; Buddenhagen et al., 2020; Ghanizadeh & Harrington, 2021). Increased identification in New Zealand may in part, be due to proactive investigation through surveys (Harrington et al., 2016; Ghanizadeh et al., 2019; Buddenhagen et al., 2020; Ngow et al., 2020; Buddenhagen et al., 2021; Ghanizadeh, Buddenhagen, Harrington, et al., 2022). Knowledge of the nature of resistance in New Zealand has been enhanced through molecular and genetic studies (Ghanizadeh, 2015; Ghanizadeh & Harrington, 2017; Ghanizadeh et al., 2019; New Zealand Plant Protection Society, 2019; Ghanizadeh et al., 2020; Ghanizadeh, Buddenhagen, Griffiths, et al., 2022). Herbicide resistance removes a key management tool. While all potential weeds are unlikely to be resistant to all potential herbicides, the loss of one or more critical control options for serious weeds has significant implications for crop managers. Effective, affordable alternatives are required.

## **2.5 Robotics and weed management**

Smart agricultural robots are now being released to market, and more are at the early development stages (Petrovic, 2020; Pardell, 2021; Petrovic, 2022; Rispens, 2022; Papadopoulos, 2023). Oliveira et al. (2021) conducted a review of more than 60 agricultural robots of which 81% were in the research stage, and 22% were designed for weeding. Merfield (2016b) noted that many autonomous systems are “self-guided

vehicles carrying weeding tools” using mechanical weeding technology little different to that used for centuries. However, ongoing advances in mechatronics, vehicle electrification and information technologies such as artificial intelligence are combining in powerful ways to offer robotic systems with automated weed recognition and precision application (Fennimore et al., 2016; Reiser et al., 2019; Ruigrok et al., 2020; Oliveira et al., 2021) and new technologies such as laser weeders are being employed (Andreasen et al., 2022; Asscheman, 2023; Silverberg, 2023). Automation enables site-specific weed management; a system of weed identification, a weed control model, and the precision application of a control method (Christensen et al., 2009). With respect to autonomous weeding in sugar beet, eight evaluation criteria and target values were proposed for a concept selection matrix: weed control efficiency (90% target), ability to target weeds (including under crop leaves), resolution of action (control border <5 mm), work rate (10 weeds sec<sup>-1</sup> per row or 0.5 m s<sup>-1</sup> operational speed), auxiliary rate (labour to assist the tool ~18 minutes per 2 hours), energy (~2 kJ m<sup>-1</sup> row), applicability to autonomous vehicles (< 150 kg, < 1 m width and height), and material costs (€11,250) (Nørremark et al., 2006). A target of 2 kJ m<sup>-1</sup> of row for inter-row weeding of crops in 50 cm rows equates to 40 MJ ha<sup>-1</sup>, which is about twice the estimates of Coleman et al. (2019) of 19 MJ ha<sup>-1</sup> for electric spot-weeding assuming five plants m<sup>-2</sup>.

Numerous agricultural robots and weed recognition algorithms are in development and are being brought together (Bloomer, 2017b; Lottes et al., 2018; Sabzi et al., 2018; Ruigrok et al., 2020; Anken & Latsch, 2022), and some are already in the market (Bakker et al., 2010; Frasconi et al., 2014; Ziwen et al., 2015; Bawden et al., 2017; Reiser et al., 2019; Carrington, 2021; Verdant Robotics, 2022). Technology to automatically locate crop rows and enable inter-row weeding was developed in the 1990s (Hague & Tillett, 2001; Tillett et al., 2002). Further developments that recognise

plants and enable intra-row weeding by machine followed (Blasco et al., 2002; Tillett et al., 2002; Slaughter et al., 2008; Xiong et al., 2017; Reiser et al., 2019). Some systems can now discriminate between common crop plants and weeds and make selective decisions (Bloomer, 2017a; Ahmad et al., 2018; Gerhards, 2018; J. Wang et al., 2019; Wu et al., 2020; Li et al., 2021; Coleman, Bender, et al., 2022). Robotic systems deploy a range of plant destruction methods, including cultivation (Tillett et al., 2002; Reiser et al., 2019), crushing (Akerman, 2015), spot-spraying (Petrovic, 2020; Ecorobotix, 2023; SwarmFarm Robotics, 2023), electric shocks (Blasco et al., 2002; Rootwave, 2019; Malewar, 2021; Baxter, 2022), laser (Heisel & Christensen, 2000; Xiong et al., 2017; Andreasen et al., 2021; Baxter, 2022) and high-intensity light (Pardell, 2021; Koerhuis, 2022). Robots such as University of Sydney's 'Ladybird' and 'Rippa' (Bogue, 2016) and Ecorobotix 'AVO' (Petrovic, 2020) operate almost 24 hours a day entirely on batteries with solar power. The robotic arms can carry electrodes instead of spray nozzles. A similar prototype research robot demonstrated in 2001 (Blasco et al., 2002) applied 90 J shocks to each seedling treated, equating to 4.5 MJ ha<sup>-1</sup>. It reached the target of one weed s<sup>-1</sup> including recognition, movement and treatment, a rate of > 3,600 weeds h<sup>-1</sup>, suggesting a work rate of 13.9 h ha<sup>-1</sup>.

Agricultural technology in New Zealand has historically focused on increasing production and profitability across the New Zealand supply chain (Ministry for Business Innovation and Employment, 2020), with notable examples including refrigerated shipping (Stringleman & Peden, 2015), electric fencing (Peden, 2008) and grass genetics (Galbreath, 2023). Realisation that the technologies themselves are export products and services has led to the government investing in the sector and encouraging its growth (Agritech New Zealand, 2020; Ministry for Business Innovation and Employment, 2020; Invest Auckland, 2023). While some express disappointment at the

rate of agritech development and adoption, there is also evidence that New Zealand farmers rapidly adopt those technologies that add value (Agritech New Zealand, 2020; Invest Auckland, 2023). Between 2000 and 2019, the availability of technology enabled agriculture to increase labour productivity in New Zealand at a rate 27% higher than the total industry average (Ministry for Business Innovation and Employment, 2023). Adoption is moderated by the technology's cost, complexity, and convenience as well as the end-user's capacity and capability to integrate the technology into the existing farming system (Bloomer & Posthuma, 2020). A simple change in method, ideally by swapping one practice for another, can be easier to implement. A requirement to change a whole system is more difficult and less likely to occur. Both Carbon Robotics (2023) and Ecorobotix (Anken & Latsch, 2022) have launched tractor-mounted versions of what were initially autonomous-robot-carried weeding systems.

## **2.6 Evolution of weed management**

Weed control thinking constantly changes. The publication of "Silent Spring" in 1962 (Carson, 2002) stimulated awareness of the undesirable effects of chemicals (McErlich & Boydston, 2014). Recommendations to develop alternatives to over-dependence on herbicides can be influenced by economics, public concern or government regulation (Burnside, 1993). By the 1990s, weed science was moving more to integrated sustainable weed management systems (Zimdahl, 1995). Integrated weed management (IWM) blends a range of control methodologies and technologies together. Negative perceptions of pesticide use and residues continue (Koch et al., 2017), with consumers often believing that naturalness, including "less chemicals" in food, is more healthy and environmentally friendly (Hoek et al., 2017). In New Zealand's global markets, especially Europe, action plans and EU mandates are mandating a reduction in pesticide

use and promoting non-chemical methods (Melander et al., 2013; Ministry for Primary Industries, 2022).

Cultural weed control includes crop rotation, stale seedbeds and avoiding seed set. Crop rotation can simplify the application of a range of weed control measures, offers different treatment windows, and diversifies weed selection pressures, reducing the build-up of species better suited to survive any particular treatment method (Lamichhane et al., 2017). The use of fallow or stale-seedbed techniques removes any weeds that germinate, providing a competition-free start for a crop. The first Europeans to arrive in New Zealand observed that Māori gardens were weed-free. It is postulated high levels of care and long fallow periods discouraged native species, and the cooler temperatures helped eliminate imported tropical weeds (Leach, 2005). However, long fallow periods that replace crop production necessitate more land for the same overall yield, are associated with increased nitrogen leaching (Francis, 1995) and reduced soil organic matter which can further reduce crop yields in the long term (Oldfield et al., 2022). Regardless of the control system, the benefits of ensuring a critical weed-free period to protect yield are well documented (Welsh et al., 1999; Keller et al., 2014; Knezevic et al., 2017; Annu et al., 2023; Kumari et al., 2023) as is the need to understand the lethal or effective dose of any treatment in a given situation (Ascard, 1995). Problem weeds that germinate and successfully establish require management.

Awareness that weed population dynamics are highly driven by agronomic practices has seen a shift from total weed destruction to rational suppression (Froud-Williams, 2017). Indeed, some suggest the notion of a weed is merely a value judgement based on ethical and social constructs rather than a scientific one. In a post-herbicide era with integrated weed management, we might recognise crop plants, weeds and other non-crop plants that do not cause harm ‘either immediately or in the longer

term' (Merfield, 2022). A multifaceted systems-approach to management of herbicide resistant weeds such as annual ryegrass is increasingly being promoted, including reducing the weed seed bank by seed catching, delayed sowing, and pre-emergent and post-emergent herbicides (Matthews et al., 1996). The objective is to avoid resistant gene spread using “all cultural, mechanical and herbicidal options” within multiyear management plans (Norsworthy et al., 2012). Automation technologies now allow IWM to move from “broadcast applications” of different techniques to one where each plant in a field can be categorised and given custom treatments based on biology (Young et al., 2017) including herbicide resistance. For example, robotic mechanical weed control might be combined with pre- and post-emergent herbicides (Saile et al., 2022).

## **2.7 Methods of weed destruction**

Chemical weed control is relatively modern in the context of human crop production. Prior to the development of modern herbicides in the 1940s, cultural and physical weed control technologies were the only options available (Froud-Williams, 2017). The efficiency and efficacy of herbicides rapidly led to their adoption as the primary control technique, becoming increasingly dominant from about the 1960s (McErlich & Boydston, 2014). Not currently permitted in New Zealand, genetically introduced herbicide resistance in crops in countries such as the USA has increased reliance on a narrow range of herbicides, notably glyphosate, in those countries using such crops, depleting farmland biodiversity and increasing the rate of herbicide resistance development (Lamichhane et al., 2017). A wide range of herbicide types is available for weed control, including selective and non-selective products, contact herbicides, some with translocated and/or residual action, that may be applied pre-sowing, pre-emergence, or post-emergence of the crop (Ross & Childs, 1995). The

herbicide selected will depend on the species present and the growth stage of plants treated, the climatic or soil conditions and other factors. Further classification of herbicides is based on their mode of action, “the overall manner in which the herbicide affects a plant at the tissue or cellular level” (Christensen et al., 2009). The wide variety and potential for selectivity makes herbicides a powerful management tool. But while herbicides have been a proven and economically viable weed control option, the effectiveness and public acceptance of herbicides are being challenged (Datta & Knezevic, 2013). The amount of herbicide applied can be reduced using automated systems with machine vision and artificial intelligence to apply the best chemical at the optimal dose only to individual weeds (Christensen et al., 2009). However, a desire for healthier food (Wooliscroft et al., 2014), soil and ecosystems, a preference to avoid the use of chemicals, and the increasing prevalence of herbicide resistance, are driving efforts to find alternatives (Peruzzi et al., 2017; Grelet & Lang, 2021).

Non-chemical weed management strategies have been well documented (Parish, 1990; Nørremark et al., 2006; Melander et al., 2017; Peruzzi et al., 2017; Jabran & Chauhan, 2018; Merfield, 2018). They may be included in a multifaceted weed control programme that still includes herbicides. A wide range of tools has been developed since agriculture began 10,000 years ago, and variations continue to appear. Physical weed control methods that can be applied post-emergence rely on separation of shoot from root, uprooting and subsequent desiccation, burial, and/or above-ground tissue rupture (Nørremark et al., 2006). Cultivation by ploughing, discing, harrows, tines, or hoes to separate, uproot or bury weeds also damages soil structure and may have a high labour and energy cost (Kaufman & Schaffner, 1982; Laguë & Khelifi, 2001; Ascard et al., 2007; Balzhaeuser et al., 2012; Coleman et al., 2019). The disturbance of soil may disperse and bury seed, induce dormancy, and promote additional weed germination

(Vleeshouwers & Kropff, 2000; Tørresen et al., 2017). Non-soil-engaging methods rely on the destruction of above-ground parts by mowing (Donald, 2007; An et al., 2020), abrasion (Forcella, 2009b; Forcella et al., 2011; Perez-Ruiz et al., 2018; Wortman et al., 2018; Forcella, 2019), crushing (Akerman, 2015) or seed destruction at harvest (Korres et al., 2019). Removing above-ground parts may not kill all weeds but can reduce competition and prevent seeding.

Thermal controls include the use of flame, hot air, steam, hot water, radiation, and electrothermal equipment. Microwaves (Whatley et al., 1973; Höschle, 1984; Sartorato et al., 2006; Brodie et al., 2007; Kacan et al., 2018), ultraviolet light (Khalilov & Akhmedov, 1992; Andreasen et al., 1999; Knezevic et al., 2016), laser pyrolysis (Heisel et al., 2001; Griepentrog et al., 2006; Rakhmatulin & Andreasen, 2020) and high-intensity light (Johnson et al., 1989; Koerhuis, 2022) are alternative thermal technologies. Thermal weeding technologies can control weeds without mechanical contact and can operate in the presence of high crop or mulch residues. Flame and high-energy electric or laser systems have a high fire risk, especially in dry organic residue environments. For autonomous systems with no human observer in the field to respond, this is particularly important.

## **2.8 Electric weeding**

As a discipline, electric weeding has a long history, with the first patents in the 1890s (Sharp, 1893; Scheible, 1895), and it has subsequently been the subject of ongoing development and patent applications (Opp & Opp, 1952; Pluenneke, 1975; Dykes, 1979; Carr, 1997; Diprose, 2016; Rona et al., 2019). Development in the 1970s and 1980s (Dykes, 1979; Diprose & Benson, 1984) competed with the advent of cost-effective translocated herbicides such as glyphosate, and ‘weed wiper’ technology (Diprose et al., 1985). With increasing objections to glyphosate and increasing herbicide

resistance, electrical weeding is again the subject of research and development (Lati et al., 2021; Lysakov et al., 2021; Bloomer et al., 2022; Lehnhoff et al., 2022). Many technology firms are active in the field, some reviving the 1980s systems, others moving to smaller robotic-mounted systems (Vigneault & Benoît, 2001; McCool et al., 2018; Rootwave, 2019; Schneider, 2020a). The commercial electrical systems available today (Kaufman & Schaffner, 1982; Moretti, 2021) have high energy demands and are suited only to certain weed/crop scenarios. Electric weeding plant selectivity has been achieved by physical separation, notably treating taller weeds in low-growing crops, such as bolters in sugar beet (Vigneault et al., 1990).

Understanding of electro-impulse weeding is lacking in many areas, including requirements and mechanisms of plant destruction, the modes and safety of electrical equipment and optimum forms of electrodes (Judaev & Brenina, 2008; Korres et al., 2019). Relative to continuous alternating current (AC), Judaev and Brenina (2008) reported weeding with high voltage (DC) impulses requires less energy, less-bulky equipment, and increases work safety. They sought to determine “electric energy lethal doses” that caused irreversible tissue damage in a range of weeds and noted plants at the end of flowering/beginning of fruit set require more energy than at other development phases. Using 18 kHz AC, Rootwave claims effective weed control using 50 MJ ha<sup>-1</sup>, stating that the very high-frequency electricity is safer than the usual 50 Hz frequencies or DC systems (Claver, 2022).

There are two main electric weeding technology categories: continuous-contact high-energy systems and pulsed-discharge low-energy systems. Diprose et al. (1978) and Diprose and Benson (1984) investigated electrothermal weed control using continuous contact alternating high-voltage electrodes and suggested that through the plant’s resistance, electrical energy is converted to heat and the cell membranes are

disrupted by rapid heating and volatilization. In field trials with large beets, destruction was achieved with 100–200 kJ per plant applied energy. Effectiveness varied according to plant size, species, age, and soil moisture status. Extensive or large in-ground root systems may not be damaged if the electric current earths before passing through the tissue (Diprose et al., 1980). Vigneault and Benoît (2001) noted the rhizomes of couch (*Elytrigia repens*) will survive several treatments with continuous electric current. High-voltage, short-pulse, low-energy weed control is possible. Electric weeding systems that use low-energy pulsed electric shocks for selective weed management have been researched and prototypes developed (Mizuno et al., 1990; Blasco et al., 2002; Yudaev et al., 2019) but not commercialised. A 5 kV pulsed system targeting individual weeds is described by Bennett (2019), but online images suggest it may be continuous contact and the reported tractor power take-off shaft energy source implies high energy is employed. An alternative is to use high-voltage, short-duration pulsed systems, which require much less energy. Slesarev et al. (1970) published research on controlling weeds with microsecond 25 kV electric pulses. Three days after treatment, 3–4 cm tall fathen (*Chenopodium album* L.) had completely stopped respiring, transpiring, and photosynthesising. Other broadleaf plants treated also died, and the researchers reported that perennial sow thistle (*Sonchus arvensis* L.) roots were killed to a depth of 23 cm. Mizuno et al. (1990) report Russian techniques using 30–50 kV DC discharges or repeated 30–80 kV pulses of over 100 J. For safety and energy efficiency, they recommended reducing the discharge energy by pulsing charges.

Mizuno et al. (1990) developed apparatus for laboratory use and showed small plants (40–60 mm height, 1–3 mm stem diameter) could be destroyed by one spark discharge of 0.14 J energy and very large plants (800–1,200 mm height, 10–15 mm stem diameter) with repeated pulses totalling only 2 J. A low-energy spot-weeder

incorporating a number of safety features was reported by Mizuno et al. (1993) to kill *Poa annua* L. in golf courses. Powered by a 12 V battery, the 3 kV 200 W rated system used high-frequency discharge. Blasco et al. (2002) describe a robotic weeder applying 15 kV at 30 mA for about 200 ms (= 90 J) and reported successful trials. They stated electrical discharge control removes the need for weed species discrimination but provided no evidence. They reported that lettuce crop plants with more than ten leaves were unaffected, with only directly affected leaves showing any damage. Harvey et al. (2019) reviewed electric weeding and suggested that an exponential increase in required energy as plants get larger makes small plant management desirable. Their unpublished report described a prototype electric system able to supply very short high-voltage pulsed discharges. Applying 6.5 J or 20 J to both broadleaf weeds and grasses (~ 6 tillers, leaf length < 10cm), they observed no immediate signs of impact but recorded plant deaths several days later. While effective in laboratory testing, the system was not further developed.

My own early studies (Chapter 3) applying pulsed electric microshocks showed small broadleaved weeds, and to a lesser extent grasses can be controlled with very small energy requirements (Bloomer et al., 2022). Laboratory trial results showed 5 J was sufficient to kill or severely limit the growth of many seedlings up to 15 cm height. This is as little as 1% of the energy of, and more effective than, ultralow energy treatments reported in recent research (Lati et al., 2021; Lehnhoff et al., 2022). To control herbicide-resistant weeds at five plants  $\text{m}^{-2}$ , the required energy would be about  $0.25 \text{ MJ ha}^{-1}$  plus transport and actuation energy for weed destruction, as compared to an optimum target of about  $20\text{--}40 \text{ MJ ha}^{-1}$  including transport suggested by Nørremark et al. (2006). My system uses pulsed DC electricity which is safer than AC because the “let go of parts gripped” is less difficult and the threshold of ventricular fibrillation is

considerably higher for shock durations longer than the period of the cardiac cycle (Standards Australia/Standards New Zealand, 2022). In reviewing the safety of conducted electrical weapons such as TASER®, Panescu et al. (2017) noted that relevant international standards that specify safety requirements for electrical medical devices and electrical fences “give very relevant guidance”. However, like my pulsed electric shock weeder, conducted electrical weapons use much higher frequency pulses, so an alternative is needed. My research apparatus has pauses between pulses so the capacitors can fully recharge. I typically have  $< 20 \text{ pulses s}^{-1}$  at  $< 0.2 \text{ J pulse}^{-1}$ , so the energy is below the 5 J limit. A commercial device should seek to increase pulse frequency for work-rate efficiency and may increase pulse energy, so the safety factor must be addressed.

While my early studies were mostly undertaken in a greenhouse, the field application of pulsed electric microshocks was later shown to be viable (Chapters 4 and 5) as a manual option or, with advances in automation, robotics and image analysis, it would be a viable precision agritech opportunity as described by Blasco et al. (2002). The energy demand is potentially highly competitive with any existing or proposed weed control option, but knowledge of the correct dose and treatment point is very limited. Experimental devices were built in Soviet countries (Yudaev et al., 2019) and Japan (Mizuno et al., 1990) in the 1970s, but no evidence of commercialisation has been found.

Combining a targeted low-energy system with the automation and vision technologies now available promises an energy and cost-efficient method of non-herbicide weed management, particularly important with the increasing emergence of herbicide resistance. Such systems can be modular, with small units combinable to cover wide swaths as required. A system killing weeds with 2 J electric “doses”

(Mizuno et al., 1990) could use perhaps 4 MJ ha<sup>-1</sup> at 100 weed seedlings m<sup>-2</sup>, equivalent to the lowest energy requirement of any system reported by Coleman et al. (2019) and an order of magnitude lower than their estimates for site-specific electric weeding at five plants m<sup>-2</sup>. Energy demand would be several orders of magnitude lower than thermal weeding techniques.

Open-source software and hardware designs such as Open Weed Locator and open image-libraries empower community development of intelligent systems (Coleman, Salter, et al., 2022). This is important for vegetable and arable producers who grow niche crops, usually missed by proprietary commercial products.

## **2.9 Energy demands of weeding systems**

The energy requirements of different weeding methods have been variously determined and reported (Kaufman & Schaffner, 1982; Barber & Lucock, 2006; Barber, 2010; Coleman et al., 2019). Comparisons of total energy requirements for broadcast weed control suggest values for light mechanical techniques (e.g. flexible tines, basket weeders) of 4–17 MJ ha<sup>-1</sup>. Mowing which ranges from 30–285 MJ ha<sup>-1</sup>, is about an order of magnitude higher, and ploughing which ranges from 614–768 MJ ha<sup>-1</sup> is higher again (Coleman et al., 2019). Herbicide control is variously reported as 15 MJ ha<sup>-1</sup> (Coleman et al., 2019), 38–115 MJ ha<sup>-1</sup> (Audsley, 2000), and 127 MJ ha<sup>-1</sup> (Alluvione et al., 2011). However Helsel and Pimentel (2007) note that herbicides have a very high energy demand in manufacture which adds considerably to the overall energy demand. They provide examples ranging from 9 MJ ha<sup>-1</sup> for chlorsulfuron, 567 MJ ha<sup>-1</sup> for glyphosate, to 880 MJ ha<sup>-1</sup> for propanil at recommended application rates. Systems involving heat from flame, air, steam, or radiation typically have very high energy demands in a range of 1,000–4,000 MJ ha<sup>-1</sup>. De Cauwer et al. (2015) tested hot water weed control in bagged plants and reported applied energy intensities as high as 39,000

MJ ha<sup>-1</sup> without achieving full control. Microwave systems were reported to use between 10,000 and 75,000 MJ ha<sup>-1</sup> (Coleman et al., 2019) as a broadcast application.

Highly targeted laser weeding of very small plants at the cotyledon stage may require 50–125 J plant<sup>-1</sup> (2.5–6 MJ ha<sup>-1</sup>), although much higher energy requirements are reported depending on wavelength and plant size (Wöltjen et al., 2008; Kaielerle et al., 2013; Andreasen et al., 2022). Coleman et al. (2021) laser treated ryegrass (*Lolium rigidum* Gaudin) and found 93% of three-leaf plants were controlled at 75 J plant<sup>-1</sup> with 300 J plant<sup>-1</sup> sufficient to control plants with up to seven leaves. However, the electro-optical efficiency of CO<sub>2</sub> lasers is only about 10% (Wöltjen et al., 2008) so the input energy is ten times the dose delivered. This suggests energy demands of about 25–150 MJ ha<sup>-1</sup> for small plants at five plants m<sup>-2</sup> or 100–600 MJ ha<sup>-1</sup> applying a broadcast treatment with 200,000 plants ha<sup>-1</sup>. Weeding systems that involve weed and crop recognition and autonomous control and application also have a high energy cost for processing and actuation. System specific details are not available, although the Carbon Robotics system uses 21 NVidia GPUs (Ward, 2023), 12 high resolution cameras, 9 LED lightbars and controls for 30 lasers (Carbon Robotics, 2023). The system is carried and powered by a large tractor so the total energy may be in the vicinity of 900 MJ ha<sup>-1</sup>. This corresponds to about 26 L diesel ha<sup>-1</sup>, or 15.5 L hr<sup>-1</sup> which is reasonable.

Electrothermal weed control is much more efficient than flame, air, or steam, which have very low heat transfer efficiencies (Merfield, 2016a). Even so, the energy requirements reported for electrothermal weed control have a very wide range, with values of 0.04 to > 200 kJ plant<sup>-1</sup>, depending on species and age (Vigneault et al., 1990). Treating individual plants, using a scenario of five plants m<sup>-2</sup>, which is a reasonable assumption for weed populations surviving other control attempts (Kaufman &

Schaffner, 1982; Vigneault et al., 1990), this equates to anywhere between 2 and 10,000 MJ ha<sup>-1</sup> for the direct energy cost, to which must be added the transport of the weeding equipment around the field. A newly developed system using very high-frequency AC was reported to give effective broadcast control using 50–100 MJ ha<sup>-1</sup> (No-Till Farmer, 2022). Coleman et al. (2019) report continuous contact electrocution of two-leaf-stage broadleaf weeds at five plants m<sup>-2</sup> requires 19 MJ ha<sup>-1</sup> and spark electrocution 14.5 MJ ha<sup>-1</sup>, values at the lower end of their calculations. Estimates for grasses are not given. My own studies applying pulsed microshocks showed small broadleaf weeds, and to a lesser extent grasses, can be controlled with very small energy requirements of about 5 J per weed (Bloomer et al., 2022) which equates to about 0.25 MJ ha<sup>-1</sup>, nett of transport and actuation energy. By comparison, Blasco et al. (2002) reported using 4.5 MJ ha<sup>-1</sup> nett of transport and actuation. A solar powered system requires virtually no extra external energy, so my system could operate at about 1% of the 20–40 MJ ha<sup>-1</sup> target for autonomous weeding proposed by Nørremark et al. (2006).

Relative energy requirements of some weeding systems are summarised in Table 2-1.

Where conversion from diesel to MJ equivalents was required, diesel consumer energy of 38.4 MJ L<sup>-1</sup> was used rather than the primary energy value of 46.3 MJ L<sup>-1</sup> reported by Barber and Stenning (2022).

Table 2-1 Estimated energy requirement ranges of selected selective and broadacre weeding methods.

<b>Weeding Method</b>	<b>Low Energy Estimate MJ ha<sup>-1</sup></b>	<b>High Energy Estimate MJ ha<sup>-1</sup></b>
<b>Selective (Site Specific)</b>		
Pulsed microshocks <sup>1</sup>	0.25	
Pulsed electric <sup>2</sup>	4.5	
Herbicides – site specific <sup>3</sup>	15	
Electrocution – site specific <sup>3</sup>	15	19
Hoeing – site specific <sup>3</sup>	17	
Laser pyrolysis – site specific <sup>3</sup>	15	249
<b>Broadacre (Whole field)</b>		
Flextine harrow <sup>3</sup>	4	6
Spoon weeder <sup>3</sup>	10	11
Herbicide <sup>4</sup>	38	115
Herbicide including embodied energy <sup>3</sup>	46	712
Herbicide including embodied energy <sup>5</sup>	47	1,007
Laser pyrolysis <sup>6</sup>	100	900
Electrocution <sup>7</sup>	32	77
Power harrow <sup>8</sup>	230	384
Hot water <sup>3</sup>	1,500	7,600
Hot water <sup>9</sup>	3,840	19,150
Microwave <sup>10, 11</sup>	9,600 <sup>10</sup>	75,000 <sup>11</sup>

1. Bloomer et al. (2022)
2. Blasco et al. (2002)
3. Coleman et al. (2019)
4. Kaufman and Schaffner (1982); Audsley (2000)
5. Calculated from Audsley (2000), Hesel and Pimentel (2007) and Alluvione et al. (2011)
6. Estimated from Wöltjen et al. (2008), Coleman et al. (2021) and Carbon Robotics (2023)
7. Kaufman and Schaffner (1982)
8. Calculated from Barber (2010)
9. Estimated from Martelloni et al. (2021)
10. Brodie (2012)
11. Calculated from Wayland et al. (1975), Hesel and Pimentel (2007) and Alluvione et al. (2011)

## 2.10 Where to from here?

Cultural weed control, including crop rotation, stale seedbeds and avoiding seed set, can form the foundation of an integrated weed management system. Crop rotation can simplify the application of a range of weed control measures, offers different treatment windows, and diversifies weed selection pressures, reducing the build-up of species better suited to survive any particular treatment method. Within a multi-year, multifaceted systems approach, the overall operations mix for effective, sustainable weed suppression can allow for occasional higher-cost systems, particularly if they effectively manage the seed bank and enable other cheaper methods to be continued. If problem weeds do germinate and successfully establish, they must be managed. High weed densities are best managed using methods such as stale seedbeds, perhaps using broadcast herbicides or light cultivation. Dealing with lower weed populations, especially herbicide-resistant escape weeds, those that emerge late or that emerge through cover mulches, and populations in sensitive crops, may be more efficiently managed using automated systems and precise application.

Preference to avoid applying agrichemicals to food and the increasing prevalence of herbicide resistance can remove herbicides as tools. Precision guidance allows for inter-row cultivation in bare-soil situations but can stimulate more weed germination. Conservation or regenerative vegetable and arable systems with an emphasis on retaining soil cover require weeding equipment that operates effectively in high residue conditions. Mechanical weed control is difficult, and hand-labour is expensive and hard to obtain. To manage costs and reduce climate impacts, low-energy systems are preferred. As a component of integrated weed management, pulsed electric microshocks as I have researched meet these criteria with a fraction of the energy required by any comparable system. Deployed as an automated weed identification and

control system, my method offers an ultralow-energy, non-contact option well suited to higher residue, non-herbicide vegetable or arable production systems such as organics, conservation agriculture, or regenerative agriculture. The system can be deployed by fully automated, solar-powered, field-robots, or as modular units on booms carried by conventional tractors to cover wide swaths rapidly. Appropriate dose rates, delivery specifications, and real-time assessment of effective treatment of weeds need further investigation, and best application within an integrated weed management system requires refinement. Different species are not equally susceptible to microshocks, so a shift in weed species spectrum could be expected if this was the only control technique used.

# Chapter 3. Proof of Concept

MICRO ELECTRIC SHOCKS CAN CONTROL BROADLEAVED AND GRASS WEEDS

## 3.1 Publication

This chapter was published as:

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This proof of concept research focused on weed control efficiency and energy as elements of a system that would include machine vision and robotics to control escape weeds in field crops. Two pulse generation systems, one single and one multiple, were developed and evaluated at different delivered voltages and energies. Greenhouse trials showed precisely applied, direct current (DC) microshocks with precisely controlled voltage, pulse number, pulse length and period can kill small plants with minimal energy. Plants took as much as two weeks to die. My results showed that 5 J is sufficient energy to bring about death or severe growth limitation in many seedlings up to 15 cm height. This is as little as 1% of the energy of, and more effective than, ultra-low energy treatments reported in other research.

**Statements of Contribution:** See Appendix 1 Statements of Contribution

## 3.2 Abstract

A search for energy efficient, non-herbicide weed control methods led to development of a novel electrical weeding technology. This study focuses on weed control efficiency and energy as elements of a system that would include machine vision and robotics to control escape weeds in field crops. Two pulse generation systems, one single and one multiple, were developed and evaluated at different delivered voltages and energies. Greenhouse trials using specially designed and built application and recording technology showed the application of precisely applied microshocks with precisely controlled direct current (DC) voltage, pulse number, pulse length and period (hereafter PMS) can kill small *Lolium multiflorum* Lam., *Chenopodium album* L., *Amaranthus powellii* S. Wats. and *Solanum nigrum* L. plants with minimal energy. Plants took as much as two weeks to die. Increasing applied energy increased effectiveness as determined by plant biomass reduction and death rate. Grasses appear difficult to control once tillering has commenced, and high voltages may destroy leaf blades but not growing points. Broadleaved plants took several days to show evidence of chlorosis which preceded senescence and death. My results showed that 5 J is sufficient energy to bring about death or severe growth limitation in many seedlings up to 15 cm height. This is as little as 1% of the energy of, and more effective than, other reported ultra-low energy treatments. To control five herbicide resistant weeds  $\text{m}^{-2}$ , the required energy would be about 0.25 MJ  $\text{ha}^{-1}$  plus transport and actuation energy for weed destruction, as compared to an optimum target of about 20–40 MJ  $\text{ha}^{-1}$  including transport suggested in the literature. PMS can effectively control broad-leaved weed seedlings and small non-tillering grasses at a fraction of the energy required by commercially available systems. This indicates PMS

has potential as a viable technology for hand-held electric weeders or as part of a site-specific robotic weeding system.

**Keywords:** nonchemical weed-control; site-specific weed management; electric weeding; robotic weeding; electric shocks; applied energy; senescence.

### 3.3 Introduction

Replacements for weed control chemicals are sought, driven by increasing herbicide resistance (Ghanizadeh & Harrington, 2020; Heap, 2020; Ngow et al., 2020), consumer preferences for chemical free food (Magnusson et al., 2003; Forbes et al., 2009; Crinnion, 2010; Hoek et al., 2017; Koch et al., 2017; Galati et al., 2019), public concern about residues in water (Bajwa et al., 2015; Hageman et al., 2019), and awareness of potential impacts of chemical treatments on soil microflora (Helander et al., 2018). A concurrent movement is towards more conservation agriculture, systems that are typically reliant on herbicides (Melander et al., 2013; Reiser et al., 2019). Conservation agriculture is accompanied by greater quantities of ground cover in the form of crops, cover crops or crop residues. This makes mechanical weed control by tillage difficult, so non-contact methods are preferable (Bauer et al., 2020).

Labour supply shortages coupled with rapid advances in robotics and vision analytics are helping push automation and systems adaptable to a robotic platform. With respect to autonomous weeding in sugarbeet, eight evaluation criteria and target values were proposed for a concept selection matrix: weed control efficiency (90% target), ability to target weeds (including under crop leaves), resolution of action (control border <5 mm), work rate (10 weeds sec<sup>-1</sup> per row or 0.5 m s<sup>-1</sup> operational speed), auxiliary rate (labour to assist the tool ~18 minutes per 2 hours), energy (~2 kJ m<sup>-1</sup> row), applicability to autonomous vehicles (<150 kg, <1 m width and height), and material costs (€11,250) (Nørremark et al., 2006). My current electric weeding research considers weed control efficiency and energy of precisely applied microshocks (PMS). The system investigated can, in combination with machine vision and robotics, address the remaining criteria.

Electric weeding has a long history, with the first patents in the 1890s (Sharp, 1893; Scheible, 1895) and has subsequently been the subject of ongoing development and patent applications (Opp & Opp, 1952; Pluenneke, 1975; Dykes, 1979; Carr, 1997; Diprose, 2016; Rona et al., 2019). Development in the 1970s and 1980s (Dykes, 1979; Diprose & Benson, 1984) competed with the advent of cost-effective translocated herbicides such as glyphosate and “weed wiper” technology (Diprose et al., 1985). With increasing objections to glyphosate and increasing herbicide resistance, electrical weeding is again the subject of research and development (Lati et al., 2021; Lysakov et al., 2021; Lehnhoff et al., 2022). Electric weeding plant selectivity has been achieved by physical separation, notably treating taller weeds in low-growing crops, such as bolters in sugar beet (Vigneault et al., 1990). Many technology firms are active in the field, some reviving the 1980s systems, others moving to smaller robotic mounted systems (Vigneault & Benoît, 2001; McCool et al., 2018; Rootwave, 2019; Schneider, 2020b). Considerable research effort is developing vision systems and plant recognition algorithms to discriminate between crop plants and weeds (Ahmad et al., 2018; Gerhards, 2018; A. Wang et al., 2019; Li et al., 2021; Coleman, Salter, et al., 2022). An electric system would be useful in wet soil conditions when mechanical weeding is difficult or ineffective. It could replace herbicides in organic food production or amenity or urban settings.

The energy requirements of different weeding methods have been variously determined and reported (Kaufman & Schaffner, 1982; Barber & Lucock, 2006; Barber, 2010). Nørremark et al. propose a target of 2.01 kJ m<sup>-1</sup> of row for inter-row weeding of crops in 50 cm rows (Nørremark et al., 2006). This equates to 40.2 MJ ha<sup>-1</sup> which is about twice Coleman et al.’s estimates of 19 MJ ha<sup>-1</sup> assuming 5 plants m<sup>-2</sup>, a reasonable assumption for post-weeding escapes (Kaufman & Schaffner, 1982; Vigneault et al., 1990).

Use of both alternating and direct current (AC and DC) electrical circuitry and spark or continuous contact applications has been reported (Judaev & Brenina, 2008; Lati et al., 2021). Most commercially available equipment uses continuous contact application, operating at tens of kilovolts, and many amps (Vigoureux, 1981; Diprose & Benson, 1984; Diprose, 2016; Merfield, 2016b). This is effective but does pose a high risk to operators or accidental contacts. Treatment at these energy levels is typified by generation of steam and sparks or flame. In dry conditions there would be a high risk of fire, a factor that must be managed. Energy of 4 kJ – 111 kJ plant<sup>-1</sup> was reported using continuous contact in greenhouse conditions (Diprose et al., 1978). To reach the target of Nørremark et al. (Nørremark et al., 2006), this would allow for only one weed every 2 – 55 m of row length. An alternative to continuous current is some form of pulsed or spark treatment. Pulsed DC currents were shown to be more energy efficient than sinusoidal AC in creating intracellular plant tissue damage (Baev & Yudaev, 2017). For the same input energy, the energy release in tissues was more than five times higher from pulsed DC.

A low-energy electrical weeding approach reported a short 15 kV AC pulse could “effectively destroy” a small *Cerastium holosteoides* plant with 304 mJ, considerably less than other systems (Mizuno et al., 1990). Observing that transpiration was reduced, it was suggested that vascular bundles were destroyed. Repeating pulses reduced electrical resistance in the plants, suggested cell wall damage increased path conductivity. However, these data were obtained from very small (0.5 g, 50 mm tall, 2 mm stem diameter) plants treated on a bench, then transferred to an agar dish. Although plants were observed to be withered at 3 days post-treatment, they were not followed through to definite death. Larger *Rumex japonicus* plants 50 cm tall withered after being treated with 30 x 2 J pulses, but again their final death is not recorded. Together these reports indicate

pulsed DC shocks might control 5 small weeds m<sup>-2</sup> with less than 5 kJ ha<sup>-1</sup>, and 5 large weeds m<sup>-2</sup> with less than 600 kJ ha<sup>-1</sup>.

Hand-held weeding devices are used for removing low population-density weeds in amenity horticulture and those remaining after broad-acre herbicide treatments. A hand-held electric weeder was reported to successfully control *Poa annua* in golf courses (Mizuno et al., 1993). The device applied treatments ranging from 450 J to 1,000 J plant<sup>-1</sup> which equates to 22.5 – 50 MJ ha<sup>-1</sup> using Coleman's calculations of 5 plants m<sup>-2</sup>, or 2 – 4 plants m<sup>-1</sup> of row to reach the target of Nørremark et al. Treatment times ranged from 1 – 3 seconds per plant, suggesting 14 – 40 hours ha<sup>-1</sup> at 5 plants m<sup>-2</sup>. While there are many applications for hand-held weeding devices, there does not appear to have been any commercialisation of the technologies described, there is little evidence of further research or development, and associated patents and applications have expired (Kimura & Mizuno, 1992; Ejima et al., 1997).

Recent results from “low energy electro-physical treatment” of weeds using two systems, one 40 kV DC and one 2.2 kV AC, showed significant reductions in weed biomass, and in some cases plant death, with smaller plants of treated species being more affected by the electric treatments (Lati et al., 2021). A range of 40 kV DC treatments applied 560 J of energy by spark or direct contact to leaf or stem of *Amaranthus retroflexus* L., *Portulaca oleracea* L., *Sorghum halepense* L. and *Citrullus lanatus* Thunb. Only direct leaf contact had significant biomass reduction on *A. retroflexus* and no treatments achieved 100% kill rate. Direct leaf contact and a stem-spark treatment significantly reduced biomass of *P. oleracea* but no treatments affected *S. halepense* or *C. lanatus*. Very small (size not stated) *Trifolium pratense* plants, 2 weeks after seeding (WAS), were

killed by the 2.2 kV AC current applying as little as 9 J but required 180 J to achieve 95% biomass reduction at 4 WAS. These doses required 1 s (9 J), 10 s (90 J) and 20 s (180 J). This implies field treatment times of about 14 – 280 h ha<sup>-1</sup> (~0.6 – 11.6 days ha<sup>-1</sup>) at 5 plants m<sup>-2</sup>. Long-duration, low-current DC electricity was also used to kill a range of small *Tamarix* spp., *Morus alba* and *Ulmus pumila* shrubs and trees and *Convolvulus arvensis* vines by Lehnhoff et al. (2022).

The focus of my research is efficient weed control with minimum energy requirements, achievable by application of precisely applied microshocks with precisely controlled direct current (DC) voltage, pulse number, pulse length and period (hereafter PMS), to deliver low energy electric treatments. This would make a simple battery-powered hand-held weeding system or high efficiency strip-weeders possible. In conjunction with the other factors outlined by Nørremark et al. (2006) it would be suitable for deployment on low-energy autonomous field robots. It would address issues of herbicide resistance, consumer desire for chemical-free food, and avoid chemical residues in water.

Here I report results from preliminary greenhouse trials of ultra-low energy weeding using PMS applied to *Lolium multiflorum* Lam., *Chenopodium album* L., *Amaranthus powellii* S. Wats., and *Solanum nigrum* L. weed seedlings. To kill plants of apparently the same size, my treatments used approximately 1% of the energy of the recently reported low-energy DC electro-physical weed treatment method. Ultra-low energy electric weed control has not been extensively studied but has numerous potential benefits. The objectives of this study were to determine a threshold “dose” of voltage and energy for mortality, and to compare relative responses of different species. Observations,

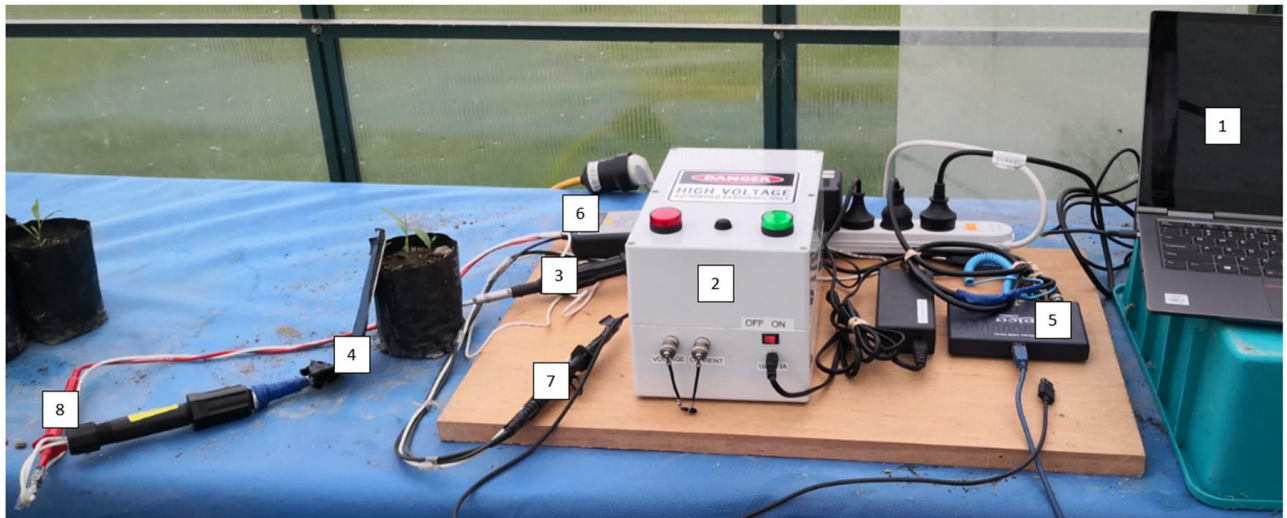
findings, and hypotheses as to the mechanism or mechanisms of plant death will direct further research to refine applications and enable a commercially viable product.

## **3.4 Materials and Methods**

### **3.4.1 Equipment and Materials**

#### **3.4.1.1 Application equipment**

Two bespoke PMS devices (Weda Tech, Hastings, New Zealand) were developed which generated DC pulses of up to 10 kV. The first developed device could deliver a single pulse of either 6 kV or 10 kV, releasing approximately 1.8 J and 5 J, respectively. The second device produced multiple pulses at voltages up to 7 kV, with pulse duration, pulse period, and either number of pulses or total delivered energy able to be programmed (Figure 3-1). The multi-pulse device was controlled using custom software running on a laptop. Each device included two 10 mm diameter aluminium rod electrodes; the earth was inserted 50 mm into the soil about 30–40 mm from the plant being treated and the positive electrode was held beside or on the stem of the plant. During the application of multi-pulse PMS, each application was measured with voltage, resistance and current being logged and energy calculated. To determine the energy absorption split between the plant and the soil, later trials included a PicoScope 2000 (Pico Technology, St Neots, Cambridgeshire, U.K.) series oscilloscope coupled with a Pico TA044 high voltage differential probe connected to the base of the plant and the earth electrode. To achieve



- |   |  |
|---|--|
| 1 Laptop with control software                          | 5 Pico Technology oscilloscope         |
| 2 Weda Tech high voltage generator and pulse controller | 6 Pico high voltage differential probe |
| 3 Earth electrode                                       | 7 Pico probe earth                     |
| 4 Treatment (positive) electrode                        | 8 Pico probe positive                  |

Figure 3-1 Photograph showing multi-pulse DC treatment equipment including picoscope to monitor voltage and current discharge.

stable voltage throughout each application, a 50 ms interval was maintained between pulses. Total treatment times varied between 0.7 ms and 3000 ms across all trials, but active times were less than 6 ms. An electronic voltage calibration check showed strong agreement between the multi-pulse device and picoscope calculated energies ( $y = 1.0273x - 0.505$ ,  $R^2 = 0.984$ ).

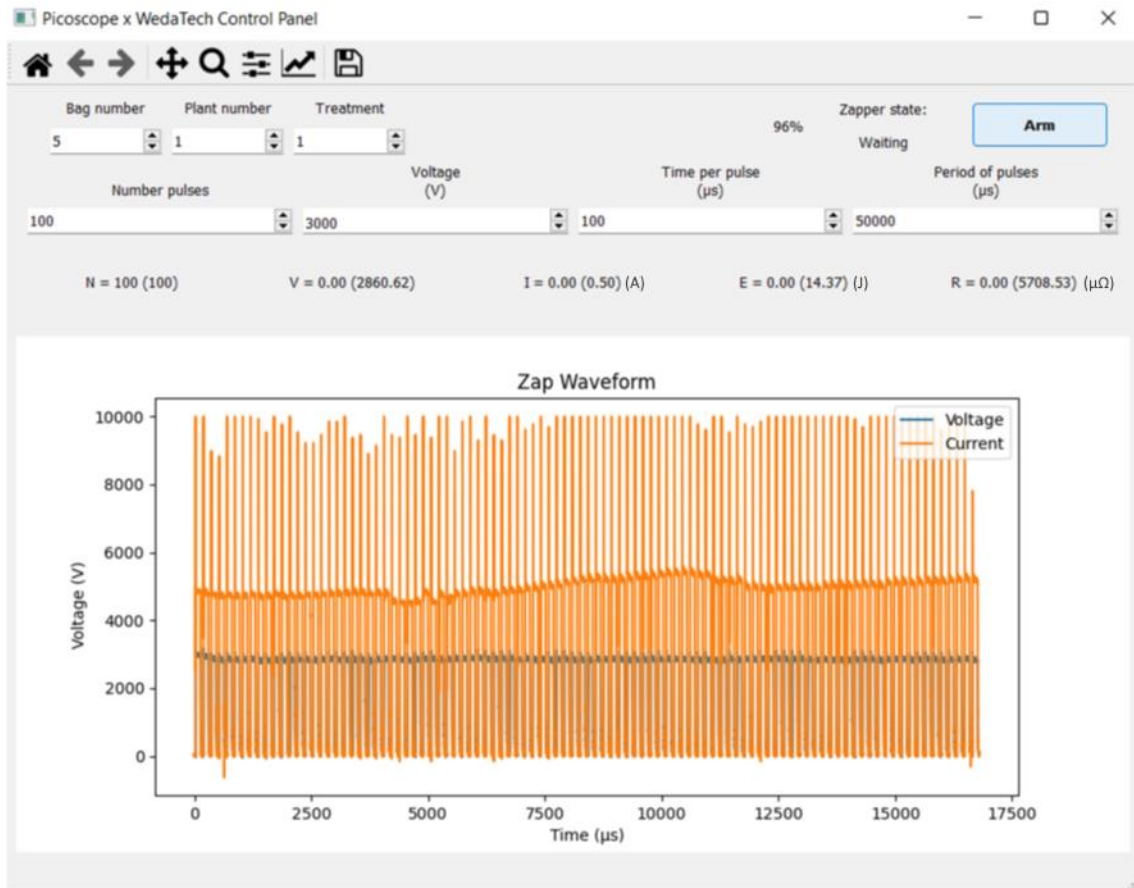


Figure 3-2 Screenshot of the Weda Tech Zapper control screen shows controllable microshock parameters, oscilloscope trace, and measured and calculated electrical values. Voltage in is volts, current is in Amperes. Calculated values are energy in Joules and resistance in microOhms.

The application and monitoring equipment set voltage, pulse number, duration and period and measured the voltage, pulse number, active time and current. Electrical power is calculated from the standard formula:

$$P = V \times I \quad \text{Equation 3-1}$$

where P is power (watts), V is voltage, and I is current (amps).

Energy transferred is calculated using the formula:

$$E = P \times t \quad \text{Equation 3-2}$$

where E = energy (joules), P is power, and t is time (seconds).

Resistance is calculated using the standard formula:

$$R = V \div I \quad \text{Equation 3-3}$$

where R is resistance (ohms), V is voltage, and I is current.

A series of uncontrolled preliminary tests identified doses that gave apparent plant-responses around which treatments were designed. I noted that in many cases few if any symptoms were evident for about 6 days, and it could be several weeks before plant death was incontrovertible. Therefore, I followed the progress of treated plants until I had confidence plants had died (fully wilted, chlorotic, roots and shoots naturally separated) or were growing healthily even if delayed relative to controls.

#### **3.4.1.2 Plant material and growing method**

In all trials the growing medium was Hastings silt loam soil sieved to 5 mm. Plants were grown in 450 mL polythene bags, placed in a greenhouse and hand watered as required. Temperature and humidity were logged and showed no values considered likely to cause any negative effect. Bags were arranged in blocks and randomised within each block.

Certified *Lolium multiflorum* cv. “Winter Star” seeds were sown six seeds to a bag and 12 – 14 days after emergence were thinned to three plants per bag, aiming to keep all plants within a similar size profile, typically with two leaves. *Chenopodium album* L. plants (20-30 mm high) were collected from wasteland, transplanted three per bag, and grown for a further 14 days before treatment. *Amaranthus powellii* S. Wats. and *Solanum nigrum* L. seedlings were collected from fallowed cropping areas. Thirty seedlings of each were transplanted one per bag and allowed to establish for 10 and 16 days before treatment.

### 3.4.1.3 Plant measurements

Prior to treatment, *L. multiflorum* plants were observed for visual differences and the number of turgid green leaves and tillers, and length of the longest leaf were measured. Following treatment, measurements were repeated, and plant deaths were recorded 15 days after treatment (DAT). Severely shrivelled or dead parts of leaf were discounted. For the multi-pulse equipment, surviving plants were measured and harvested at 15 DAT, and their fresh mass was determined. Plants from each bag were aggregated, oven dried at 62°C for 3 days and reweighed. The mean plant dry weight was determined for the surviving plants in each bag.

No relative growth measurements of *C. album* plants were made. The binary measure of effectiveness at 28 DAT was plant vitality, defined as dead or alive. *Amaranthus powellii* plants were monitored for 11 DAT recording the height to top growing point ( $\pm 1$  mm), stem diameter at 2 cm above ground ( $\pm 0.01$  mm), and a leaf area estimation ( $\pm 0.01$  cm<sup>2</sup>) using the “Easy Leaf Area Free” (ELA Free) smartphone application (Easlon & Bloom, 2014). At 11 DAT, plant status was assessed and assigned to one of three categories: healthy, collapsed or dead. For the binary logit analyses, all the plants not obviously dead were classified as alive. When *S. nigrum* plants were treated, in addition to recording machine energy discharge, the energy absorption by soil or plants was measured with the Picoscope by attaching one oscilloscope electrode to either machine electrode and the other at the base of the plant. Plants were monitored for 30 DAT recording the stem diameter ( $\pm 0.01$  mm) at 3 cm above ground, and a leaf area estimation ( $\pm 0.01$  cm<sup>2</sup>) using ELA Free. At 30 DAT plant vitality was assessed.

## 3.4.2 Treatments

### 3.4.2.1 Experiment 1: Single pulses

Single pulse treatments at 6 kV and 10 kV were applied to *L. multiflorum* and *C. album* plants. The earth electrode was set 50 mm into the soil 30 – 60 mm from the plant base. The positive electrode was placed < 5 mm from a *L. multiflorum* leaf 60 mm above the soil and < 5 mm from the *C. album* stem 50 mm above the soil. The *L. multiflorum* experiment had three plants per bag, and the *C. album* experiment had one plant per bag, with four bags per treatment and the control (Table 3-1).

Table 3-1 Trial summary table showing trial number, weed species, plant size, single electric pulse treatments applied and number of replicates.

Trial	Species	Size	Treatments	Reps	Notes
1	<i>L. multiflorum</i>	1 tiller, 2 leaves, long leaf 109 mm	Single pulse: 6 kV (1.8 J), 10 kV (5.0 J)	4	Single pulses to single leaves 60 mm above soil
2	<i>C. album</i>	50-70 mm high	Single pulse: 6 kV (1.8 J), 10 kV (5.0 J)	4	Treatment electrode <5 mm from stem, 50 mm above soil

### 3.4.2.2 Experiment 2: Multiple pulses

Multi-pulse treatments were applied at 3 kV and 6 kV, with the positive electrode held against the plant. For *L. multiflorum*, the tip was set at the top of the basal sheath about 30 mm above the soil. For *A. powellii* and *S. nigrum* the tip was 50 mm above the soil. The *L. multiflorum* had three plants in each of four bags per treatment plus the control. The *A. powellii* and *S. nigrum* had one plant in each of six bags per treatment plus control (Table 3-2).

Table 3-2 Trial summary table showing trial number, weed species, plant size, multiple electric pulse treatments applied and number of replicates.

<b>Trial</b>	<b>Species</b>	<b>Size</b>	<b>Treatments</b>	<b>Reps</b>	<b>Notes</b>
3	<i>L. multiflorum</i>	Most 1 tiller, 2 leaves, long leaf 149 mm	16 treatments: combinations of 3 and 6 kV, 2 pulse lengths, target energy 1, 3, 5 & 7 J	4	Multiple pulses at top of basal sheath about 30 mm above the soil; 3 kV pulse lengths 4 times 6 kV pulse length for equivalent energy/pulse
4	<i>A. powellii</i>	50-70 mm high	3 kV, target energy levels 1, 3, 5 and 7 J	6	Multiple pulses, treatment electrode set against stem 30 mm above soil
5	<i>S. nigrum</i>	110 mm tall, stem 4 mm diameter	3 kV, target energy levels 1, 3, 5 and 7 J	6	Multiple pulses, treatment electrode set against stem 30 mm above soil

### 3.4.3 Statistical analysis

Following the recommendations of Armstrong (2014) and Perneger (1998), no multiple test (e.g. Bonferroni) corrections were applied. For *L. multiflorum*, biomass data were analysed with each bag of three grasses representing one replicate (n=4), a sample size satisfactory for Kruskal-Wallis (Field, 2013).

The dichotomous result “Dead” or “Not Dead” at the end of each experiment was analysed using binary logistic regression. Key independent factors considered for *L. multiflorum* modelling included set voltage, set energy, energy applied, soil moisture at treatment, and total leaf length. For *A. powellii* and *S. nigrum* the modelling included set voltage, set energy, energy applied, leaf number, shoot number and stem diameter.

Those parameters that did not add to the model were ignored. The *C. album* plants were assessed only for turgor and erectness. For the binary logit analysis of *L. multiflorum*, each individual plant was coded dead or alive and treated as a replicate (n=12).

## **3.5 Results**

### **3.5.1 Experiment 1: Single pulses**

When *L. multiflorum* plants were treated with single 6 kV and 10 kV pulses (Trial 1), the fine leaves rapidly collapsed. In a few cases there was a smell of hot grass, and in one case steam was noted. At 1 DAT, all treated leaves were wilted and had turned grey. The stem below the leaf collar was normal except for four 10 kV treated plants which had wilted to ground level, and eventually, three of these four plants died. No control plants died, showed signs of damage, or of senescence. Out of 12 plants treated with 6 and 10 kV, only two and four plants, respectively, died by the end of the trial. Model coefficients (Chi-square, df and Sig. p), model summary (Nagelkerke R Square), percent correct prediction classification (cut value 0.500) and independent variables' statistical significance Sig. and odds ratio Exp(B) for the trials are presented in Table 3-3.

When *C. album* plants were given single spark pulses (Trial 2), they slowly collapsed to lie prostrate within 30 seconds of treatment. After 24 hours, 5 of 24 treated plants had regained turgor. At 28 DAT, those plants that recovered by 1 DAT were alive, and all but 2 of the remaining 19 plants were dead. All control plants survived and continued to grow. The treated plants showed high mortality with 9 of 12 plants treated with 6 kV and 8 of 12 plants treated with 10 kV dead by 28 DAT. Set voltage was a significant predictor of the likelihood of plants being killed by single pulse treatments, although this is confounded because the higher voltages also imparted much higher energy (Table 3-3)

Table 3-3 Statistical summary table of binary logistic regression analyses of factors significantly predicting the probability of *Lolium multiflorum* and *Chenopodium album* weeds dying after receiving single pulse electric treatments

Trial	Species (dose type)	Chi-square	df	Sig. p	Nagelkerke R square	Correct Case Classifn	Predictive Variable	Sig.	Exp(B)
1	<i>L. multiflorum</i> (single pulse)	12.258	3	0.007	0.486	86.1	Set Voltage	0.039	1.561
							Total Leaf Length	0.051	0.608
2	<i>C. album</i> (single pulse)	10.312	1	0.001	0.338	69.4	Set Voltage	0.011	1.000

Single-pulse treatments applied to *L. multiflorum* did not reliably kill all target plants but compared to controls, the Independent Samples Kruskal-Wallis Test showed that at higher energies the survivors generally had decreased biomass 15 DAT as expressed by longest leaf length, and total green leaf length (Table 3-4). In each case the significant difference was between the control and the 10 kV treatment.

Table 3-4 Statistical Summary of Independent Samples Kruskal-Wallis Test for a range of biometric variables measured on *Lolium multiflorum* plants 15 days after treatment.

Trial	Species (dose type)	Independent Variable	Dependent Variable	Tot n	Kruskal -Wallis Ha	df	Asymp. Sig.
1	<i>L. multiflorum</i> (single pulse)	Voltage	Leaf Number	12	5.115	2	0.077
			Tiller Number	12	3.520	2	0.172
			Longest Leaf Length	12	7.284	2	0.026
			Total Green Leaf Length	12	8.000	2	0.018
			Number of Alive Plants	12	4.015	2	0.134

The test statistic is adjusted for ties.

### 3.5.2 Experiment 2: Multiple pulses

The multi-pulse equipment allowed lower energy pulses to apply the same total energy. When multiple pulse treatments were applied to *L. multiflorum* (Trial 3), all plants immediately but slowly collapsed, lying prostrate across the bags. All the treated plants and none of the control plants exhibited signs of wilting 1 DAT. Four DAT, 48% of treated plants exhibited leaf deformation with a concertina effect in tissue that had been just inside the basal sheath when treatments were applied (Figure 3-3). The effect remained on plants that did not die (Figure 3-4). In some cases, emerging leaf tips were trapped in adjacent leaves, forming hoops as they elongated.



Figure 3-3 Close-up of “concertina” leaf deformation in *Lolium multiflorum* plants 4 days after treatment with 5 J at 6 kV.



Figure 3-4 *Lolium multiflorum* plants treated with 5 J at 6 kV shows characteristic leaf deformation still present 28 days after treatment.

The differences in total leaf number, tiller number, total leaf length, final dry-mass, and number of plants per bag that were alive 15 DAT between untreated control and treated plants were significant in all cases ( $p < 0.001$ ) according to the Independent-Samples Kruskal-Wallis test. Post-hoc pairwise comparisons showed three homogenous subsets of treatments for which the number of live plants 15 DAT was not significantly different. The 1 J treatment at 3 kV and the 1 J, 3 J and 5 J treatments at 6 kV were not significantly

different to the control or each other. There was no significant difference in number of plants that were alive 15 DAT given 3 J at 3 kV or 7 J at 6 kV, or between those given 5 J or 7 J at 3 kV. Similar results with more separation were found for total plant dry mass at 15 DAT (Table 3-5).

Table 3-5 Summary of treatments by applied voltage and energy on *Lolium multiflorum* plant dry mass and number of live plants 15 days after treatment. Groups with the same letter in a column are homogenous subsets at  $p=0.05$  according to Kruskal-Wallis post-hoc tests of pairwise comparisons.

<b>Treatment</b>	<b>Plant Dry Mass per Bag (g)</b>	<b>Number of Live Plants per Bag</b>
0-0	0.607 <sup>a</sup>	3.00 <sup>a</sup>
3000-1	0.460 <sup>ab</sup>	3.00 <sup>a</sup>
3000-3	0.074 <sup>c</sup>	0.75 <sup>b</sup>
3000-5	0.000 <sup>d</sup>	0.00 <sup>c</sup>
3000-7	0.000 <sup>d</sup>	0.00 <sup>c</sup>
6000-1	0.494 <sup>a</sup>	3.00 <sup>a</sup>
6000-3	0.319 <sup>b</sup>	2.50 <sup>a</sup>
6000-5	0.300 <sup>b</sup>	2.88 <sup>a</sup>
6000-7	0.104 <sup>c</sup>	1.25 <sup>b</sup>

Total plant dry weight 15 DAT shows the 3 kV treatments were more effective than 6 kV treatments of the same set energy level. No 6 kV treatments were fully effective, with 7 J treatment the best giving 62.5% death rate (Figure 3-5).

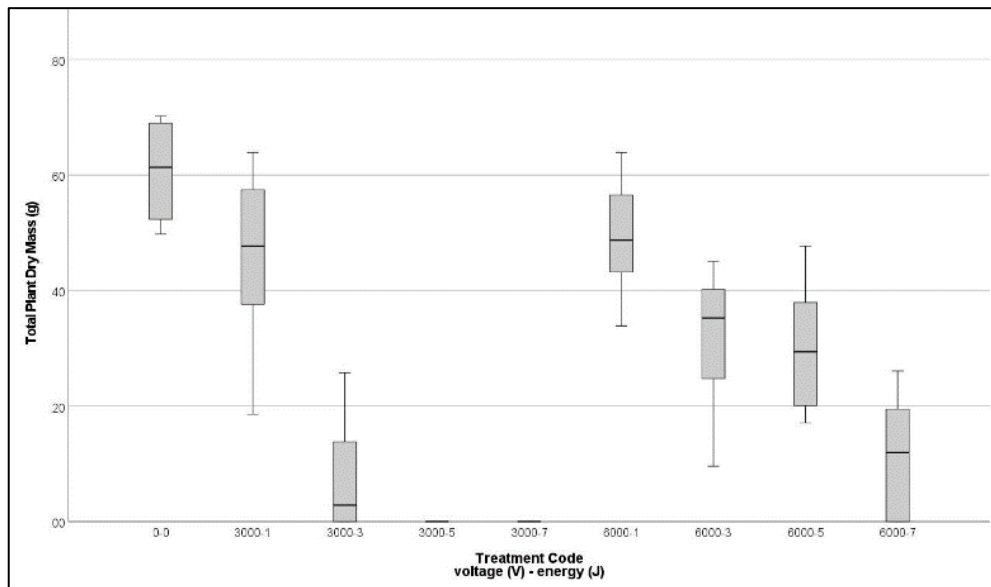


Figure 3-5 Clustered boxplot of total plant dry weight per bag of *Lolium multiflorum* 15 days after treatment by applied voltage and target energy.

Final assessments were made 15 DAT when plants were either visibly alive and growing, or dead. Among the 3 kV treatments, 75% of plants receiving 3 J and all plants receiving 5 and 7 J died. No control plants died. Binary logit analysis showed a highly significant contribution of voltage, but the odds ratio  $\text{Exp}(B)$  indicates almost no change in effectiveness with voltage increase (Figure 3-6, 3a). Investigation showed  $\text{Exp}(B)$  for voltage is confounded with the 0 kV control, because the 6 kV treatment was less effective than 3 kV. Analysis of only the treated plants, showed voltage to be a significant factor, with increasing voltage reducing the probability of death (Table 3-6, Figure 3-6, 3b).

Table 3-6 Statistical summary table of binary logistic regression analyses of factors significantly predicting the probability of *Lolium multiflorum*, *Amaranthus powellii* and *Solanum nigrum* weeds dying after receiving multiple pulse electric treatments.

Trial	Species (dose type)	Chi-square	df	Sig. p	Nagelkerke Rsquare	Correct Case Classification	Predictive Variable	Sig.	Exp (B)
3a	<i>L. multiflorum</i> (multi-pulse)	159.43	3	< 0.001	0.730	90.7	Applied Voltage	< 0.001	0.999
							Target Energy	< 0.001	4.348
							Leaf Number x Longest Leaf Length	0.028	0.897
3b	<i>L. multiflorum</i> (multi-pulse) Treated Only	147.25	2	< 0.001	0.717	89.1	Applied Voltage	< 0.001	0.199
4	<i>A. powellii</i> (multi-pulse)	9.180	2	0.010	0.354	80.0	Target Energy	0.034	1.477
5	<i>S. nigrum</i> (multi-pulse)	8.565	2	0.014	0.345	70.0	Stem Diameter	0.092	0.133
							Target Energy	0.017	1.596

When applied to *L. multiflorum* plants, delivered energy at 3 kV was about 90% of the set energy ( $r^2 = 0.97$ ) but at 6 kV only 71% of set energy was delivered ( $R^2 = 0.93$ ). Thus the 7 J at 6 kV treatment only delivered about 5 J (Figure 3-6).

All treated *A. powellii* plants (Trial 4) slowly collapsed after PMS were applied, except one plant treated with 1 J and one treated with 7 J which both remained erect. The 1 J plant had drooped 1 DAT but remained alive throughout. Another 1 J plant that initially drooped had recovered at 4 DAT and was healthy at the end of the trial. That plant absorbed half the energy of the rest of the 1 J treated plants. The erect 7 J plant remained healthy throughout. Electrical data show it received only a quarter of the mean energy of the rest of that treatment (Figure 3-7).

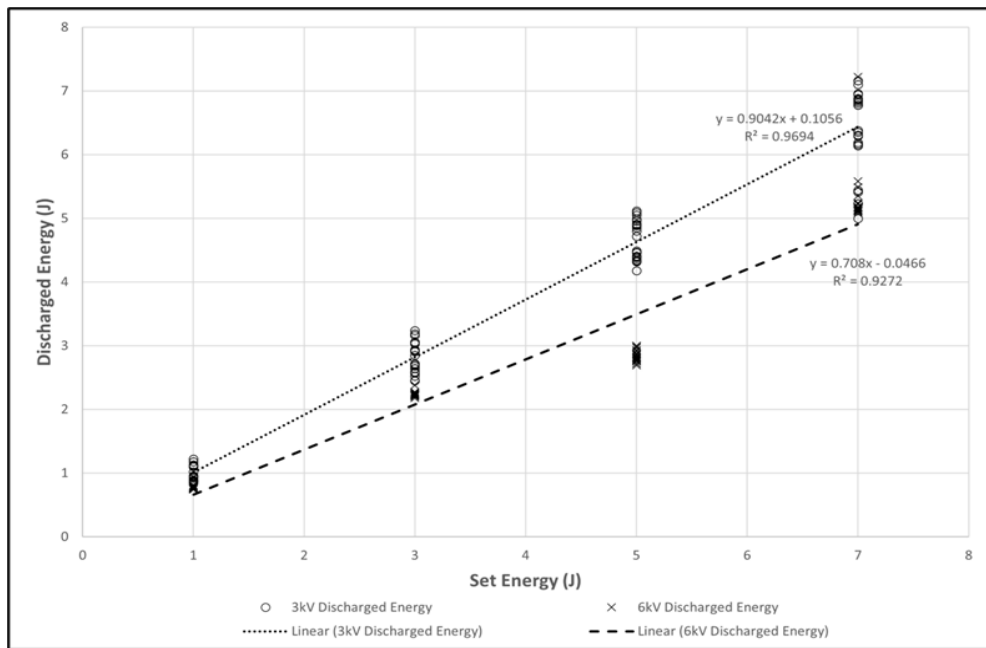


Figure 3-6 Scatterplot of equipment set energy versus actual discharged energy by voltage applied to *Lolium multiflorum*.

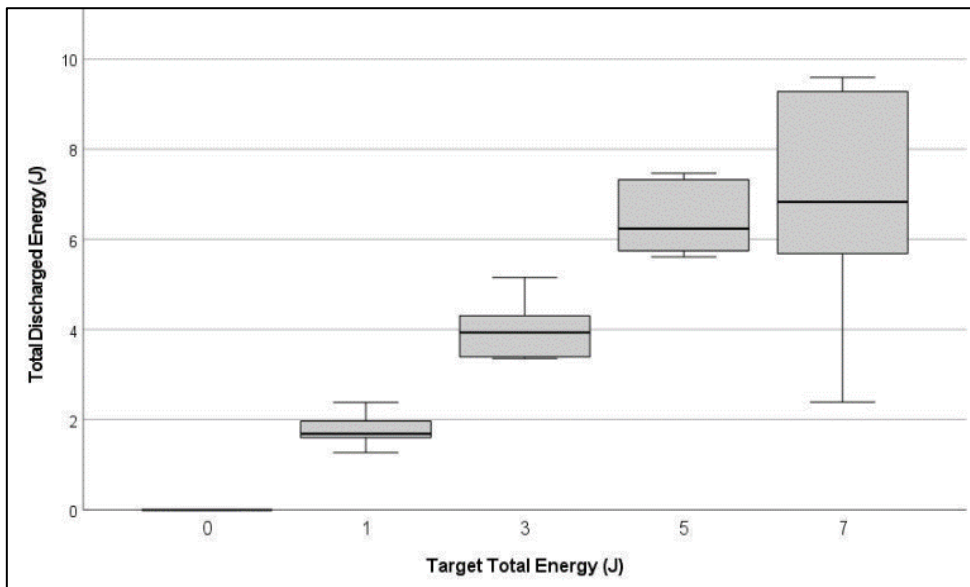


Figure 3-7 Box plot of total discharged energy by target discharged energy show higher variance among the 7 J treated *Amaranthus powellii* plants.

Throughout the trial, all *A. powellii* control plants remained erect and healthy and continued to grow. At 4 DAT, control plants had grown with height (166%), stem diameter (143%) and leaf area (215%) of size at treatment. Treated plants had decreased in size, with aggregated average results of height (62%), stem diameter (55%) and ELA leaf area (29%) of size at treatment. At 4 DAT, plants that collapsed after treatment showed severe chlorosis and wilting, with chlorosis beginning in interveinal leaf areas towards the tip of the leaf and spreading back over time to the stem to affect all leaves entirely (Figure 3-8).



Figure 3-8 *Amaranthus powellii* leaf showing typical chlorosis following PMS treatment.

At 11 DAT, the treatments were significantly different to the control, but not to each other, for stem diameter and ELA leaf area score (Table 3-7). At 11 DAT some plants in every treated set had died and only two of 24 were healthy. One of healthy plants had received only 25%, and one 48% of the energy of the rest of their treatment group. Even though some dead plants retained sections of green stem tissue, the stems had collapsed, and the hypocotyl region had decayed allowing the above ground parts to separate from roots with less than 10 mm of root attached.

Table 3-7 Statistical Summary of Independent Samples Kruskal-Wallis Test for a range of biometric factors of *Amaranthus powellii* and *Solanum nigrum* plants 15 days after receiving multiple pulse electric treatments.

Trial	Species (dose type)	Independent Variable	Dependent Variable	Total n	Kruskal-Wallis H	df	Asymp Sig.
4	<i>A. powellii</i> (multi-pulse)	Set Total Energy	Stem Diameter 11DAT	6	23.517	4	<0.001
			ELA Score 11 DAT	6	21.477	4	<0.001
5	<i>S. nigrum</i> (multi-pulse)	Set Total Energy	Stem Diameter 30 DAT	6	16.163	4	0.003
			ELA Score 30 DAT	6	15.166	4	0.004
			Fresh Mass 30 DAT	6	17.250	4	0.002
			Dry Mass 30 DAT	6	11.677	4	0.020

When *S. nigrum* plants were treated (Trial 5), the picoscope was connected between the plant base and one of the treatment electrodes. Comparing this with machine-recorded energy discharged allowed the relative energy absorption by the soil and the plant to be determined. This showed 63% ( $y=0.63x + 0.06$ ,  $R^2=0.983$ ) of discharged energy was absorbed by the above ground plant, the remainder by soil and/or roots (Figure 3-9).

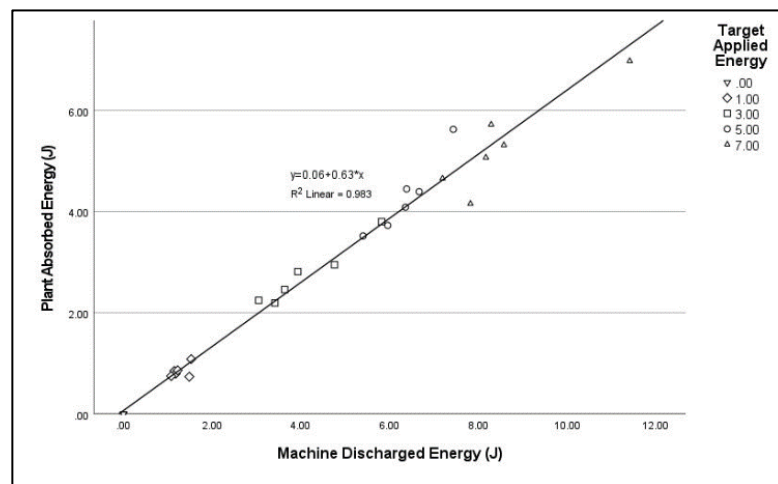


Figure 3-9 Scatterplot of energy absorbed by above-ground plant tissue when small *Solanum nigrum* plants were treated with pulsed microshocks at 3.5 kV

Control plants remained healthy and growing throughout the trial. At 8 DAT, one plant treated with 1 J and all other treated plants showed signs of main stem wilting or collapse, and curling leaves, with the degree of curling related to the energy applied, a trend that continued. At 14 DAT some treated plants remained viable with normally developing side shoots that had turgid stems and leaves. The prevalence was less as the applied energy increased. These plants were alive at the end of the trial, the rest of the treated plants were dead. At 30 DAT, final measurements were made. Treatments at 3 J or more were significantly different to the controls in all factors including final vitality, stem diameter, ELA Score and plant dry mass. Plant deaths increased as the energy applied increased, with four times more plants dead when treated with 7 J than with 1 J.

### **3.6 Discussion**

These experiments demonstrated the potential of PMS as an ultra-low energy treatment method for controlling a range of weed species at the seedling stage. Voltages were kept below 10 kV, and the energy released by the equipment was low. In general, these would place the equipment used within the criteria acceptable for electric fence energisers (International Electrotechnical Commission, 2002; Gallagher Group, 2018). The higher voltage treatments and the higher individual pulse energy treatments appeared more able to impart sufficient energy to cause immediate plasmolysis of leaf tissue in fine *L. multiflorum* seedling leaves. This is not correlated with improved plant mortality and may even reduce effectiveness. A possible explanation is that the fine leaf acts as an electrical fuse, preventing the current from impacting the rest of the plant.

The leaves of all treated *L. multiflorum* plants fell over within seconds of PMS application, indicating disruption of cell turgor. Wilting was not generally apparent in the

sheath of the plant, but deformities originating in tissues within the sheath at time of treatment were commonly observed after the leaves emerged, the concertina effect suggested some effect on expanding cells and the entrapped leaf tips suggested disruption between adjacent developing leaves. While leaf deformation in tissues inside the main sheathed shoot shows PMS affected tissue development, the survival of stems and emergence of new tillers indicated insufficient energy to kill basal growing points.

Even where *L. multiflorum* plants are not killed, ultra-low energy treatment with PMS does have a limiting effect on growth, significantly reducing growth rates for at least 2 weeks. This suggests potential to reduce competition against crop plants and may enable a strongly growing crop to get established and out-compete the grasses. There was wide variation in the growth of individual plants for any energy below 3 J. At 5 J and above all plants treated at 3 kV died, whereas many plants treated at 6 kV continued to grow, although at a reduced rate compared to the control plants. Such growth reduction may be sufficient to allow a rapidly developing crop to gain dominant competitive advantage but would not necessarily avoid weed seed bank contributions.

Several possibilities may have contributed to lower than expected energy delivery or reduced effect of a set energy level being applied. In some cases, electric arcing was observed between the positive electrode and the ground, sometimes running along the stem surface, sometimes directly through the air. This was particularly noted with 10 kV single pulses but occurred with other settings especially if the electrode was within 30 mm of the ground. In these cases, the energy discharge from the devices could be high, but the measured absorption by the plants could be very low. Multiple pulsing allows the energy of an individual pulse to be reduced but total delivered energy to be maintained.

However, in these trials, longer pulse times at higher voltages (higher energy individual pulses) could still cause arcing. In the case of high energy and high voltage pulses to fine *L. multiflorum* leaves, this was probably due to higher resistance and lower conductance, especially when leaves collapsed. When multiple pulses were applied, especially when a longer duration was required, oscilloscope measurements indicated a probable loss of direct contact for some or all the time.

There was a demonstrated effect of PMS on broadleaved weeds even at low energy levels, with treatments significantly reducing vitality and growth relative to untreated control plants. Applying these treatments to *C. album* and *A. powellii* seedlings was largely effective, and the survival of those receiving the highest target energy treatment is assumed due to poor electrode contact. Although there was some immediate effect of treatment, the results of these trials consistently show an extended period before symptoms of senescence were expressed.

Early electrical weeding research suggested that resistive heating causes boiling within the tissue, causing the cells to rupture and the plant to die (Diprose et al., 1980) although cells and vascular bundles can be damaged without boiling or burning (Eberius, 2017). The research reported here suggests for broadleaved plants especially, that a different mechanism may be involved as plants took some time to lose turgor and there was initially no external physical damage visible.

The absence of steam or odour suggests there was no gross overheating of tissues at treatment and those plants that drooped or collapsed did so slowly. The recovery of some plants after some hours indicates any membrane disruption was transitory and repairable. This suggests the effect was not caused by gross bursting of cells but indicates very porous

rather than fully ruptured membranes. The much greater diameter of the *C. album* and *A. powellii* stems compared with the *L. multiflorum* leaves is likely to have reduced electrical resistance, and therefore reduced thermal heating.

Of particular interest was the evident onset of chlorosis and senescence, and the subsequent separation of shoot and root at the hypocotyl observed in *A. powellii*. The onset and progression of chlorosis indicated that a physiological response was triggered in the treated plants. Plants may show no effect of treatment for several days to a week, and then take several weeks to die, usually expressing plant senescence. Broadleaf plant symptoms such as loss of leaf chlorophyll, browning and wilting of leaf and stems are similar to those associated with some herbicides. Various abiotic stresses have been reported to effect such a response through programmed cell death (PCD) triggered by pollutants, excessive light or UV radiation or environmental conditions including heat and cold (Vartapetian et al., 2011), leading to over-production of reactive oxygen species (ROS) (Gill & Tuteja, 2010; Sedigheh et al., 2011; Petrov et al., 2015).

No treatment killed all *S. nigrum* plants, and death was not immediate, with several weeks passing before a confident assessment could be made. These plants were notable for being very fleshy, and for having multiple shoots at time of treatment. While the treatments of 3 J or higher killed the targeted shoot in 83% of cases, in others the side shoots were not killed, or new side shoots appeared. This suggested the impact of the treatment was restricted to a section of stem and did not effectively impact the entire plant. There were significant reductions in growth associated with treatments of 3 J or over, which could potentially reduce their competitive ability against a developing crop, although this has not been assessed.

Future research repeated these trials with other species and investigate effects of changing variables such as voltage, pulse period, pulse energy and number, plant part or parts targeted, soil conditions and time of year (Chapters 4 and 5). Trials will be designed to develop better understanding of the mechanism of senescence and death.

These trials demonstrated that very low energy, high voltage pulses of direct current can kill weed seedlings with far less energy than alternative weed control options, including currently available electrical weeding systems. While these trials were conducted in a laboratory setting, with precise field application the results should be replicable. This makes the system ideal for the treatment of herbicide resistant weeds that have escaped chemical or other control methods. The small energy requirement makes the system suitable for incorporation in a mobile hand-held weeding tool, or individually or in groups in a precision robotic application.



# Chapter 4. Practical Application

PULSED MICROSHOCKS FROM A FLAT PLATE ELECTRODE ARE EFFECTIVE AT KILLING WEEDS

## 4.1 Publication

This chapter was published as:

Bloomer, D. J., Harrington, K. C., Ghanizadeh, H., & James, T. K. (2023a). The electric spatula: Killing weeds with pulsed microshocks from a flat-plate electrode. *Agronomy*, 13(11), 2694. (Bloomer et al., 2023a)

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Seeking an easy-to-deploy, energy-efficient, non-herbicide weed control method, I tested a flat-plate electrode that could be deployed via a hand-held unit or as part of a fully automated system to control escape weeds in field crops. *Lolium multiflorum* was difficult to kill but significantly reduced growth rates indicated that effective treatment of is possible. Broadleaf weeds are more susceptible. Indications are that the energy required to kill 100% of seedlings will vary from 0.1 to 1.0 MJ ha<sup>-1</sup>, easily meeting my target effectiveness and efficiency goals.

**Statements of Contribution:** See Appendix 1 Statements of Contribution

## 4.2 Abstract

Seeking an easy-to-deploy, energy-efficient, non-herbicide weed control method, I tested a flat-plate electrode to apply pulsed electric microshocks (PMS) to a grass and four broadleaf weed species. The method can be deployed via a hand-held unit or as part of a fully automated system to control escape weeds in field crops. The effectiveness of the treatments and the relative energy discharges when applying similar electric doses to the plant leaves or to the plant when pressed to the soil with a flat-plate electrode were compared. The method killed only half of the treated *Lolium multiflorum* “Winter Star” plants, well below my target rate, but significantly reduced growth rates and indicated that effective treatment of  $<1.0 \text{ MJ ha}^{-1}$  for treating five plants  $\text{m}^{-2}$  is possible. *Polygonum aviculare* L., *Amaranthus powellii* S. Wats., *Amaranthus deflexus* L., and *Solanum nitidibaccatum* Bitter plants were successfully controlled, with the energy required to kill 100% of seedlings varying from 0.1 to 0.9  $\text{MJ ha}^{-1}$ . This indicates that broadleaf weeds are more susceptible than grasses. This result easily met my target effectiveness and efficiency goals. The discharged energy increased when the electrode pressed the plant to a dry soil surface rather than to the leaves only, and increased further when the electrode pressed the plant to a wet soil surface.

**Keywords:** nonchemical weed-control; site-specific weed management; electric weeding; robotic weeding; electric shocks; applied energy; grass weeds; broadleaf weeds.

### 4.3 Introduction

For decades, herbicides have provided easy-to-use, cost-effective weed management, but now consumer preference for chemical-free food (Hoek et al., 2017; Koch et al., 2017; Galati et al., 2019), awareness of environmental impacts (Helander et al., 2018; Hageman et al., 2019), regulation increasingly restricting agrichemical use (Beckie et al., 2020; Alcántara-de la Cruz et al., 2021), and increasing prevalence of herbicide resistance (Ngow et al., 2020; Buddenhagen et al., 2022; Harrington & Ghanizadeh, 2023), are forcing changes to weed management strategies (Bloomer et al., 2023b). The emergence of agritechnologies incorporating automation, machine vision and artificial intelligence (Ruigrok et al., 2020; Oliveira et al., 2021; Ecorobotix, 2023; Papadopoulos, 2023) and development of new techniques for weed destruction (Lati et al., 2021; Bloomer et al., 2023b; Carbon Robotics, 2023) offer alternatives that minimise or avoid the requirement for herbicides, avoid soil disturbance and can work effectively in high crop or crop-residue conditions. My focus has been on electric weeding which I have previously discussed (Bloomer et al., 2022, 2023b). Slaven et al. (2023) provided a thorough review of methods and commercially available equipment.

Adopting the evaluation criteria of Nørremark et al. (2006) for autonomous weeding in sugar beet, the target weed control efficiency should be > 90%, and the energy demand < 2 kJ m<sup>-1</sup> of a 50 cm wide row, which equates to 40.2 MJ ha<sup>-1</sup>. This is about twice the 19 MJ ha<sup>-1</sup> (380 J plant<sup>-1</sup>) energy estimates assuming five plants m<sup>-2</sup> of Coleman et al. (2019), a density considered reasonable for post-weeding escapes (Kaufman & Schaffner, 1982; Vigneault et al., 1990). Most commercially available electric weeding equipment uses continuous contact alternating current (AC), operating at 5 – 15 kV and 0.5 – 2 amps (Merfield, 2016b). Energy of 4 kJ – 111 kJ plant<sup>-1</sup> was reported using continuous contact

in greenhouse conditions (Diprose et al., 1978). To reach the target of Nørremark et al. (2006), this would allow for only one weed every 2 – 55 m of row length.

The amount of energy required for effective control is related to plant size. Lati et al. (2021) showed that as little as 9 J killed very small (size not stated) *Trifolium pratense* plants using 2.2 kV AC shocks two weeks after seed sowing (WAS). However at 4 WAS, 180 J were required to achieve 95% biomass reductions. Baev and Yudaev (2017) stated that the effect on vegetation of any damaging factor is related to the energy involved. Working with weed stem fragments *in vitro*, they showed that pulsed direct current (DC) was more than five times more energy efficient than sinusoidal AC in creating intracellular plant tissue damage. Harvey et al. (2019) suggested an exponential relationship, and thus highest energy efficiency would be achieved by treating weeds when small and young.

I identified electric weeding with pulsed electric microshocks (PMS) as a very low-energy option requiring a fraction of the energy of any other system. I demonstrated in greenhouse trials that a minimal amount of electrical energy was sufficient to control small, non-tillering grass and broadleaf weed seedlings up to 15 cm in height using a precisely placed point electrode (Chapter 3). At 5 J plant<sup>-1</sup>, my system treating five plants m<sup>-2</sup> would require 0.25 MJ ha<sup>-1</sup> plus transport and actuation energy to control a range of non-tillering grasses. A simple flat electrode plate applied to plants was thought more practically realistic for field deployment as a simple hand-held weeding system, on low-energy autonomous field robots, or for application by high efficiency strip-weeders. It would simplify application, particularly to effect control of very small weed seedlings. Even with a ten-fold increase in energy, my system's energy requirement would be an

order of magnitude lower than other reported systems. Human safety needs to be considered when deploying high-voltage equipment but I note that commercially available high-voltage weeding systems use far higher energy than my PMS system. As described in Chapter 2 (Bloomer et al., 2023b), regulations do not specifically address systems such as ours, which approximates the risks associated with electric fence energisers.

This chapter presents results from greenhouse trials using PMS applied with a flat plate to a grass, *Lolium multiflorum* “Winter Star”, and four different species of broadleaf weed seedlings, *Polygonum aviculare* L. (wireweed), *Amaranthus powellii* S. Wats. (redroot), *Amaranthus deflexus* L. (prostrate amaranth), and *Solanum nitidibaccatum* Bitter (hairy nightshade). The objectives of this study were to determine if a flat plate used in a way practical for paddock-treatment could apply a threshold “dose” of voltage and energy to achieve > 90% mortality, to assess energy expenditure, and to compare relative responses of different species. Two electrode placements were tested: the plate pressed against the plant only with a 3 cm separation from the soil surface, and the plate pressing the plant on to the soil, with the soil surface either wet or dry. The effectiveness in terms of plant death or biomass reduction, and energy consumption are reported.

## **4.4 Materials and methods**

### **4.4.1 Equipment**

A custom built PMS device which produced multiple DC pulses of up to 4.5 kV was developed by Weda Tech (Hastings, New Zealand). The device was controlled using custom software running on a laptop with discharge voltage, pulse duration, pulse period, and number of pulses able to be programmed. To improve accuracy and ensure full

capacitor recharge, a 50 ms interval was maintained between pulses. A PicoScope 2000 series oscilloscope was coupled with a Pico TA044 high voltage differential probe connected to the positive and the earth electrodes to monitor applied doses. Pulse data were automatically recorded and logged in csv files.

#### **4.4.2 Plants and growing method**

In all trials, the growing medium was Hastings silt loam soil sieved to 5 mm. Plants were grown in 450 mL polythene bags, placed in a greenhouse and hand watered as required. Temperature and humidity were logged and showed no values considered likely to cause any negative effect. Bags were arranged in blocks and randomised within each block. Each trial had six replicates.

Six certified *L. multiflorum* “Winter Star” seeds were sown in each bag and 12 – 14 days after emergence, the seedlings were thinned to three plants per bag, aiming to keep all plants within a similar size profile, typically with three or four leaves. The *P. aviculare*, *A. powellii*, *A. deflexus*, and *S. nitidibaccatum* seedlings were collected from fallowed cropping areas, transplanted one per bag and allowed to establish for 16 – 21 days before treatment.

#### **4.4.3 Treatment method**

The voltage was set between 3.0 and 4.5 kV, pulse length between 25 and 5,000  $\mu$ s, and the number of pulses was selected to adjust the total energy potentially delivered by each treatment. A 5 mm diameter aluminium earth electrode was inserted into the soil at the base of the plastic bag being treated (Figure 4-1). The positive electrode was a flat aluminium plate 75 mm x 100 mm (similar in shape and size to a kitchen spatula) pressed



Figure 4-1 Application method showing the earth electrode inserted horizontally into the base of the bag and the flat plate electrode ready to be pressed against the weed.

fully to the soil surface or only to the leaves. For “leaf” treatments, the plate was set on a plastic spacer placed on the soil to maintain a gap of approximately 30 mm above the soil surface. For “soil” treatments, the plant and plate were pressed to the soil. During the application of PMS, each pulse was measured with voltage, current, pulse duration and pulse period logged, and resistance and discharged energy were calculated.

One or two days prior to treatment, the top of the soil in “dry” treatments was cultivated 1 – 2 cm deep to encourage a layer of dry soil peds at the surface. As *L. multiflorum* had three plants in each bag, the plate electrode was pressed to all plants in each bag at once so that the dose was shared among the three plants. A range of treatments was applied, with various combinations of voltage, pulse length, pulse number, electrode placement and soil surface moisture status compared against controls that were pressed but not shocked. The minimum voltage was 3 kV, and the maximum was 4.5 kV (Table 4-1).

Table 4-1 Trial summary table showing trial number, weed species, plant size, and treatments applied to *Lolium multiflorum*, *Polygonum aviculare*, *Amaranthus powellii*, *Amaranthus deflexus* and *Solanum nitidibaccatum*.

Trial	Species	Mean Size	Treatments
1	<i>L. multiflorum</i>	1.63 tillers, 3.65 leaves, longest leaf 141 mm	Control plus nine treatments: 3 kV, 100 $\mu$ s pulses, 50, 100 and 200 pulses applied to the leaves only, or leaves pressed to a dry or wet soil surface
2	<i>L. multiflorum</i>	1.91 tillers, 4.05 leaves, longest leaf 197 mm	Control plus 10 treatments: 100 x 200, 200 x 200 and 200 x 400 $\mu$ s pulses at 3.5 kV applied to leaves only or on leaves pressed to a dry soil surface; 100 x 100, 200 or 400 $\mu$ s 4.5 kV pulses, or 50 x 100 $\mu$ s 4.5 kV pulses, pressed to a dry soil surface
3	<i>P. aviculare</i>	42 mm stem length, 1.0 mm stem basal diameter, 12.2 leaves	Control plus 10 treatments: 3.0 and 4.5 kV, pulse lengths 50 – 200 $\mu$ s, 100 – 400 pulses, plate pressed to soil. One treatment of 4.5 kV, 200 x 200 $\mu$ s pulses was applied to the leaf only
4	<i>A. powellii</i>	34.3 mm stem length, 1.6 mm stem basal diameter, 8.5 leaves, 3.5 sideshoots	Control plus eight treatments with the electrode on leaves only or on leaves pressed to soil, 4.5 kV, 100 or 200 $\mu$ s pulse length, 50, 100 and 200 pulses. 1 treatment with 10 x 1000 $\mu$ s pulses
5	<i>A. powellii</i>	64.3 mm stem length, 1.8 mm stem basal diameter, 9.8 leaves, 5.8 sideshoots	Control plus 8 treatments 4.5 kV, 25 and 50 x 100 $\mu$ s pulses with electrode on leaves only or on leaves pressed to dry soil; 5 x 250, 500 or 1,000 $\mu$ s pulses, or 1 x 5000 $\mu$ s pulse pressed to dry soil
6	<i>A. deflexus</i>	40.1 mm stem length, 1.4 mm stem basal diameter, 7.2 leaves, 4.6 sideshoots	Control plus one treatment at 4.5 kV with 50 x 100 $\mu$ s pulses applied to leaves pressed to a dry soil surface
7	<i>S. nitidibaccatum</i>	63.0 mm stem length, 2.9 mm stem basal diameter, 27 leaves, 3.5 sideshoots	Control plus eight treatments at 3.18 and 4.50 kV, 25 and 50 x 100 $\mu$ s pulses with electrode on leaves only or on leaves pressed to dry soil; and 1 x 5000 $\mu$ s pulse at 3.18 kV to leaves pressed to dry soil

#### 4.4.4 Statistical analysis

Prior to treatment, *L. multiflorum* plants were visually assessed and the number of green leaves and tillers were counted, and length of the longest leaf of each plant was measured. Following treatment, measurements were repeated, and plant deaths were recorded 10 days after treatment (DAT). Severely shrivelled or dead parts of leaf were discounted.

Surviving plants were measured, harvested, and their fresh mass was determined. Plants from each bag were aggregated, oven-dried at 62°C for 3 days and reweighed. The mean plant dry weight was determined for the surviving plants in each bag.

Prior to treatment, the *P. aviculare*, *A. powellii*, *A. deflexus* and *S. nitidibaccatum* plants were measured and graded by size, before being sorted into equivalent groups for treatment. They were monitored for 24 days post-treatment, at which time final measurements were made, including survival (dead or alive), days to death, stem length, stem basal diameter, fresh mass of green tissue, and subsequent dry mass after oven drying at 62°C for 3 days. All the plants not obviously dead were classified as alive. Dead plants had no green mass and were recorded as 0 g.

Data were statistically analysed using SPSS Version 28.0.1.0 (142). Where biomass differences are reported, the default for statistical comparisons of groups was the independent samples Kruskal-Wallis one-way ANOVA (k samples) because almost all cases failed the ANOVA homogeneity of variance using Levene's test. Homogenous subsets were determined from post-hoc analyses and the stepwise step-down multiple comparisons. Following the recommendations of Armstrong (2014) and Perneger (1998), no multiple test (e.g. Bonferroni) corrections were applied. For *L. multiflorum*, biomass data were analysed with each bag of three grasses representing one replicate (n=6), a sample size satisfactory for Kruskal-Wallis (Field, 2013). The dichotomous result "dead" or "not dead" at the end of each experiment was analysed using binary logistic regression. *Polygonum aviculare*, *A. powellii* and *S. nitidibaccatum* modelling included set voltage, electrode placement, set energy, energy applied, leaf number, shoot number and stem diameter. Those parameters that did not add to the model were discarded.

## 4.5 Results

Throughout all trials, all control plants grew healthily.

### 4.5.1 *Lolium multiflorum*

When 3.0 kV multiple pulse treatments were applied to *L. multiflorum* (Trial 1), only three of 180 plants died. However, the Kruskal–Wallis test showed there were significant differences in the final plant dry mass at 10 DAT ( $H(9) = 31.64, p < 0.001$ ), with the two higher-dose treatments (100 x 100  $\mu\text{s}$  pulses and 200 x 100  $\mu\text{s}$  pulses) that pressed the plants to dry soil being the most effective (Figure 4-2).

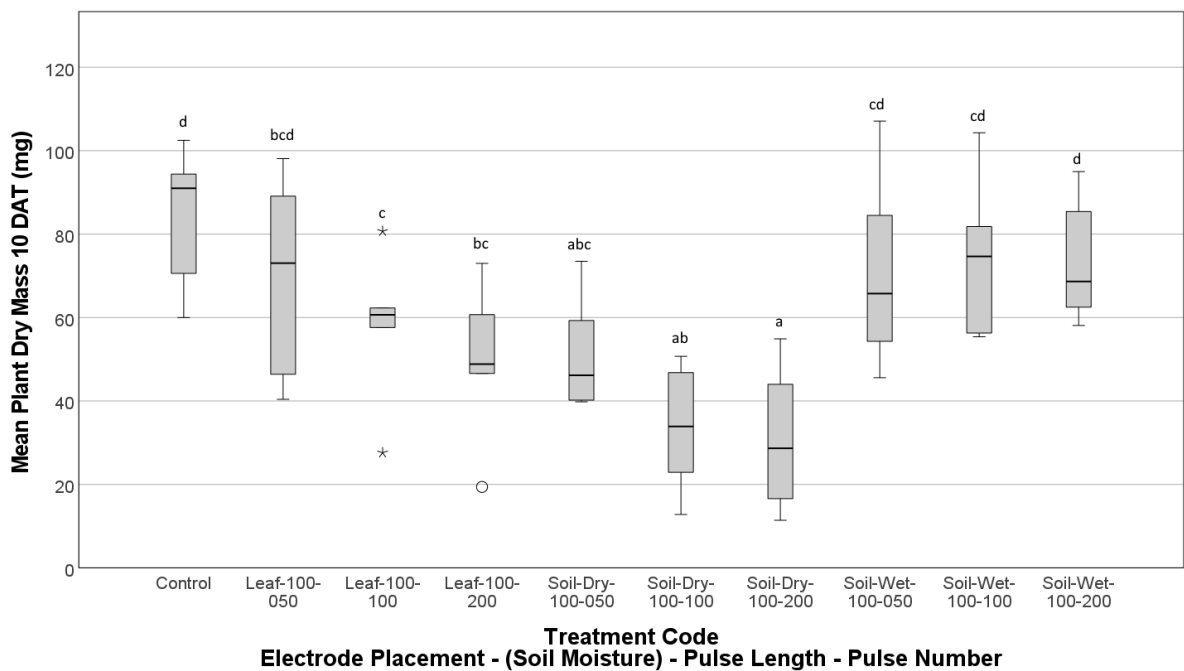


Figure 4-2 Simple box plot of mean *Lolium multiflorum* plant dry mass per bag 10 days after treatment by treatment applied with treatments varying by placement of the electrode (electrode contact), the electric pulse length ( $\mu\text{s}$ ) and the number of pulses applied in the treatment. Asterisks indicate extreme outliers, and the circle indicates a mild outlier.

Doubling the number of pulses doubled the amount of energy discharged. Excluding the untreated controls, the energy discharged under each treatment was compared using the Kruskal–Wallis test, which showed that there were significant differences ( $H(8) = 49.713, p < 0.001$ ). Treatments were categorised into homogeneous subsets based on an asymptotic significance level of  $p = 0.05$ . The same doses applied to dry soil discharged more energy than those applied to leaves only but fell into the same homogenous subsets. Applications to wet soil resulted in up to five times more energy being discharged than the same dose applied to leaves only, such that the lowest dose in the wet soil treatment discharged the same energy as the highest leaf or dry soil dose (Figure 4-3).

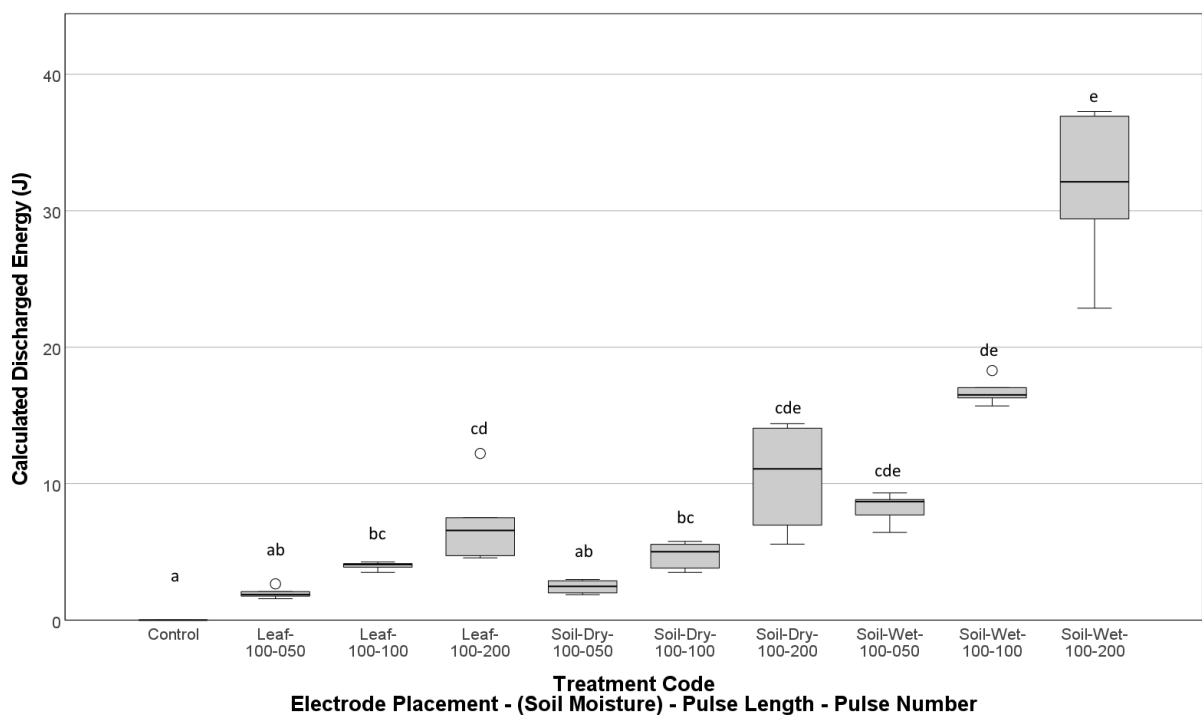


Figure 4-3 Simple boxplot of calculated discharged energy (J) by treatment when multiple DC electric pulses or varying pulse lengths were applied to sets of three of *Lolium multiflorum* seedlings in bags with a flat plate electrode pressed to the leaves only, or to the soil with either a dry or wet soil surface. Mild outliers are indicated by small circles.

On the basis of these results, a second *L. multiflorum* trial (Trial 2) was conducted at two higher voltages, 3.5 and 4.5 kV. The leaf versus dry soil surface application comparisons continued but the wet soil treatments were dropped. Of 192 plants treated in Trial 2, only 34 died. The most effective leaf-applied, 3.5 kV treatment with 200 x 400  $\mu$ s pulses killed 44% plants treated at an average of 69 J per application or 23 J plant<sup>-1</sup>. The most effective treatment was 4.5 kV treatment with 100 x 400  $\mu$ s pulses pressed to soil, which killed half the plants at an average of 102 J or 34 J plant<sup>-1</sup>.

The lowest dose leaf treatment (3.5 kV, 100 x 200  $\mu$ s pulses), the lowest dose 3.5 kV soil applied treatment (100 x 200  $\mu$ s pulses), and lowest two 4.5 kV soil applied treatments (50 x 100  $\mu$ s pulses and 100 x 100  $\mu$ s pulses) were ineffective with no plants dying (Figure 4-4).

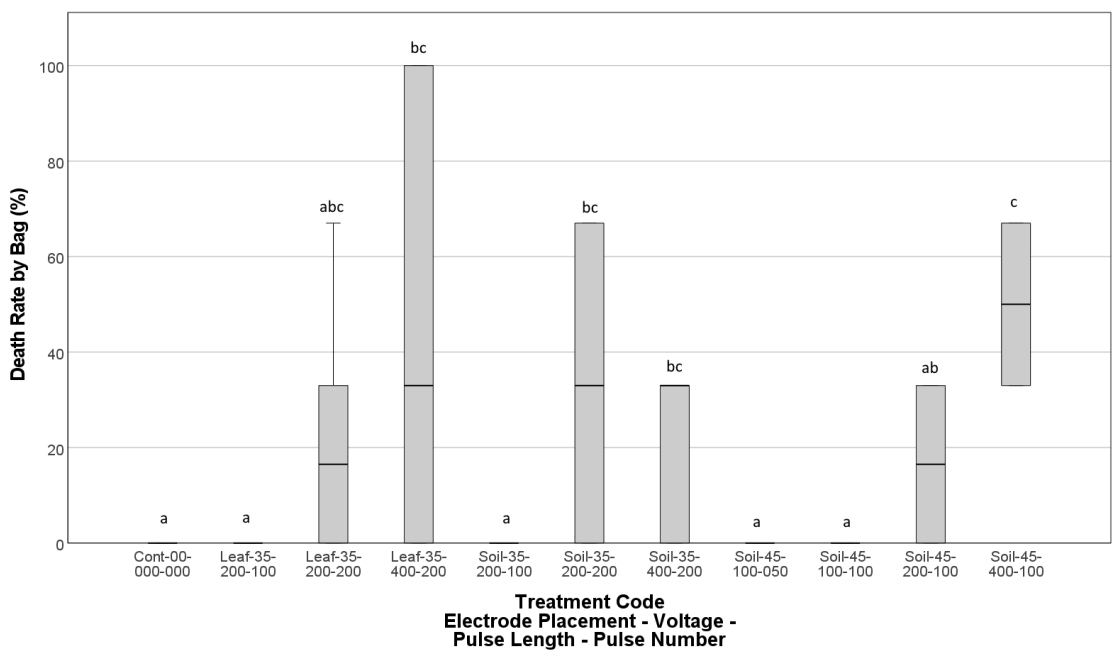


Figure 4-4 Simple box plot of *Lolium multiflorum* final death rate per bag when treated with pulsed microshocks at a range of voltages, pulse lengths and number of pulses which were applied to leaves only or to leaves pressed to the soil.

The Kruskal-Wallis test showed there were significant differences in the plant death rate by 14 DAT ( $H(10) = 35.3, p < 0.001$ ) with all treatments that resulted in any deaths being significantly different to the control as assessed by homogeneous subsets based on an asymptotic significance level of 0.05.

The Kruskal-Wallis test showed there were significant differences in the final plant dry mass (Figure 4-5) at 14 DAT ( $H(10) = 40.9, p < 0.001$ ). The leaf-applied treatment at 3.5 kV with 100 x 200  $\mu$ s pulses and the soil-applied treatment at 4.5 kV with 50 x 100  $\mu$ s pulses were not significantly different to the control. The soil-applied treatment at 3.5 kV with 100 x 200  $\mu$ s pulses and the treatment at 4.5 kV with 100 x 100  $\mu$ s pulses did not result in any plant deaths but the dry mass of plants from these treatments at 14 DAT were significantly different to the control.

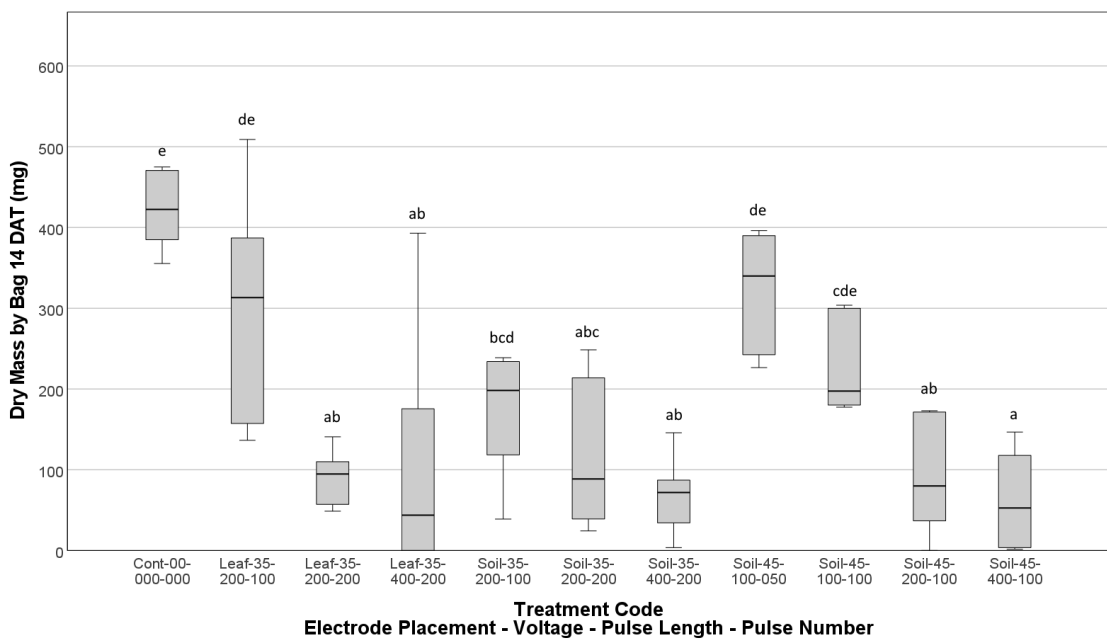


Figure 4-5 Simple box plot of *Lolium multiflorum* plant dry mass by bag (mg) measured 14 days after treatments were applied to leaves only or to leaves pressed to the soil with pulsed microshocks at 3.5 or 4.5 kV with different pulse lengths and number of pulses applied to plants.

Plotting the final plant dry mass against the energy discharged by treatments indicated a point at which plants reach zero dry mass, i.e. will be killed. Figure 4-6 shows collected data and a linear trend line, with the individual 90% confidence lines fitted.

A review of the energy discharged by treatment showed predictable increases as the voltage, length of pulses or number of pulses increased. At 3.5 kV, the energy discharge rate was 51% higher for the soil contact compared with leaf only contact. Energy discharge rate of soil contact was 93% higher at 4.5 kV than at 3.5 kV.

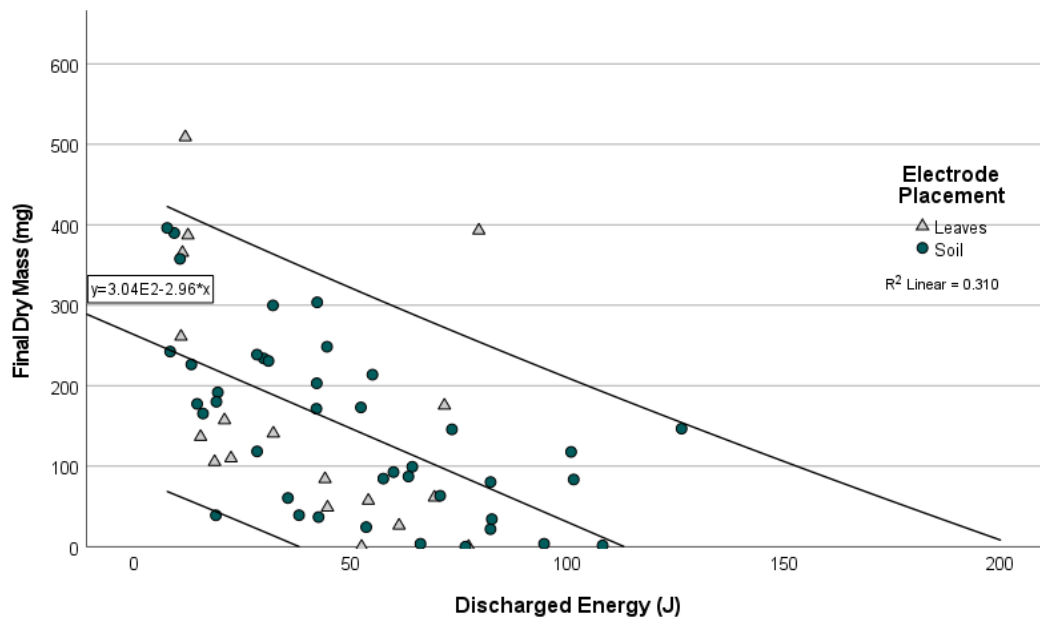


Figure 4-6 Scatter plot of *Lolium multiflorum* plant dry mass (mg) by discharged energy (J) by electrode placement with overall linear fit line bounded by 90% individual confidence levels.

#### 4.5.2 *Polygonum aviculare*

Treatments for *P. aviculare* applied both 3.0 kV and 4.5 kV doses to plants pressed to the soil surface, with pulse length and number aiming to double and redouble energy applying between 15 and 120 J. However, machine firmware limited the possible number of pulses, and the maximum was reduced to 81 J. An additional treatment applied 4.5 kV doses with the flat electrode pressed to the leaves only. Pressing the plant to the soil surface without also applying electric shocks did not kill any plants. The leaf-applied 4.5 kV treatment killed all plants (Figure 4-7) despite the discharged energy being far less than soil treatments (Figure 4-8).

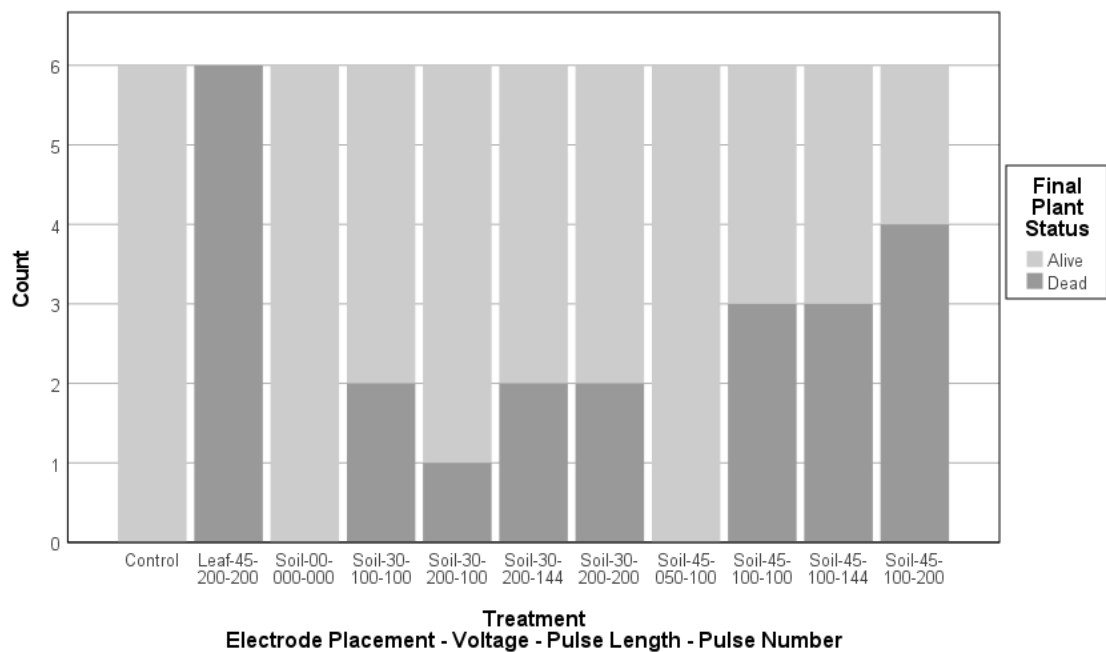


Figure 4-7 Stacked histogram count of treatment by final status of *Polygonum aviculare* plants that were subjected to treatments at 3.0 kV and 4.5 kV with different electric pulse lengths and number of pulses applied with the electrode pressed to the leaves only or to the leaves pressed against the soil.

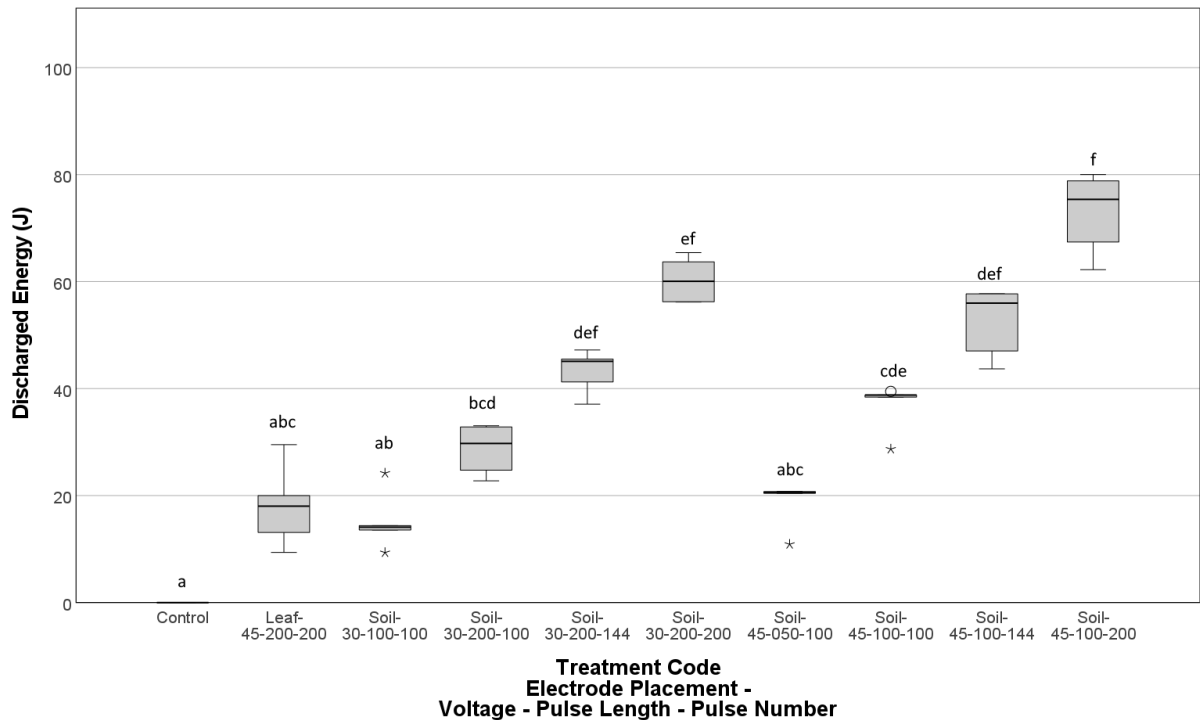


Figure 4-8 Simple box plot of discharged energy (J) applied to *Polygonum aviculare* by treatments at 3.0 kV and 4.5 kV at different electric pulse lengths and a different number of pulses applied with the electrode pressed to the leaves only or to the leaves pressed against the soil. Extreme outliers are indicated by stars. A mild outlier is indicated by a small circle. Plots sharing the same letters are not significantly different.

The average energy discharge was 17.9 J plant<sup>-1</sup>, equivalent to 0.895 MJ ha<sup>-1</sup> if treating five plants m<sup>-2</sup>. With the plate electrode pressing plants to the soil surface, about one-third of the *P. aviculare* weed seedlings were killed by the 3.0 kV treatments with no greater effect as energy increased. The 4.5 kV treatments did show an increase in effectiveness with increasing energy although no soil applied treatments were fully effective. The most effective was the strongest 4.5 kV treatment that applied 200 x 100 μs pulses with a kill rate of 66%. The same patterns were found in the final dry mass measurements (Figure 4-9). The Kruskal-Wallis test showed there were significant differences in final dry mass (H (10) = 32.29, p < 0.001) but only between the control and leaf-applied 4.5 kV, 200 x 200 μs pulses (p = 0.001), control and soil applied 4.5 kV, 200 x 100 μs pulses (p = 0.006), and control and soil-applied 4.5 kV, 144 x 100 μs pulses (p = 0.032).

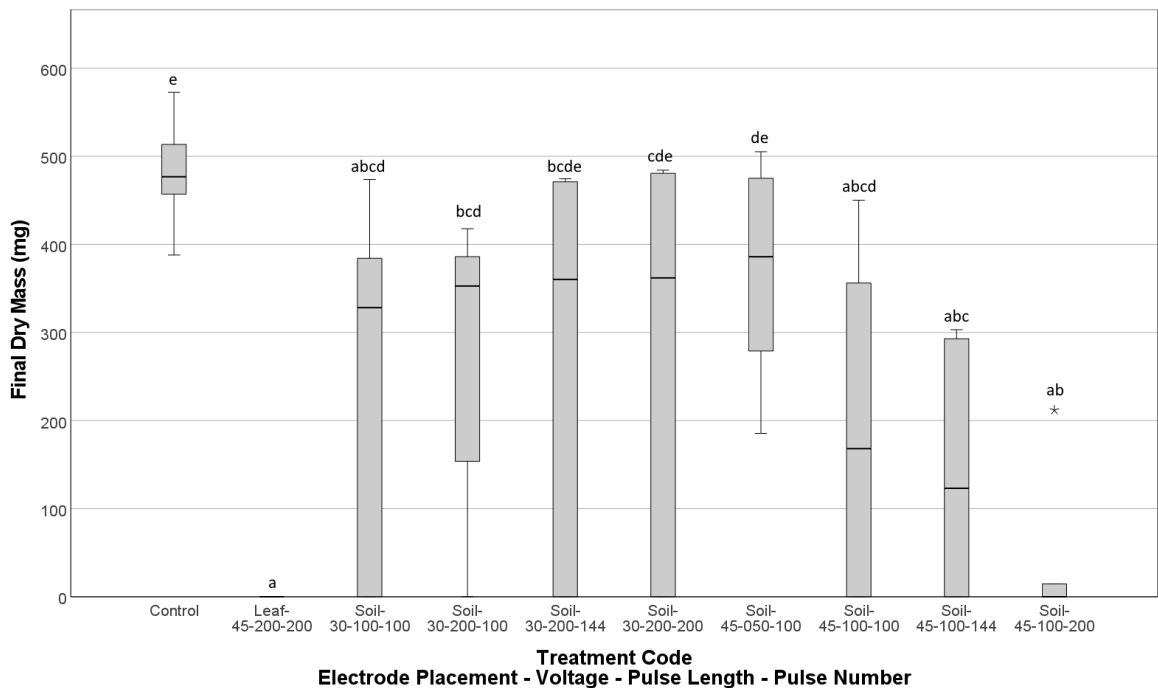


Figure 4-9 Simple boxplot of *Polygonum aviculare* final dry mass following application of treatments at 3.0 kV and 4.5 kV with different electric pulse lengths and a different number of pulses applied with the electrode pressed to the leaves only or to the leaves pressed against the soil. An extreme outlier is indicated by the star. Plots sharing the same letters are not significantly different.

A logistic regression was performed to ascertain the effects of treatment on the likelihood that plants would be killed. The logistic regression model was statistically significant,  $\chi^2(10) = 32.733$ ,  $p < 0.011$ . The model explained 53.9% (Nagelkerke  $R^2$ ) of the variance in the outcome and correctly classified 77.3% of the cases. A second regression was performed using set voltage, discharged energy, stem length, stem diameter, leaf number, soil of leaf contact and pulse length on the likelihood that plants would be killed. The logistic regression model was statistically significant,  $\chi^2(8) = 23.351$ ,  $p < 0.003$ . The model explained 43.8% (Nagelkerke  $R^2$ ) of the variance in outcome and correctly classified 76.7% of cases. Hosmer and Lemeshow also indicated good fit  $\chi^2(8) = 5.213$ ,  $p < 0.735$ . However no variables were independently significant. When the leaf application treatment was removed from analysis, the model was not statistically significant,  $\chi^2(5) =$

6.716,  $p = 0.243$ . The model explained 16.4% (Nagelkerke  $R^2$ ) of the variance in outcome but correctly classified 74.1% of the cases.

The measured discharged energy when the plate electrode was pressed to the soil was very close to values estimated based on earlier trials, with overall  $R^2 = 0.956$ . The mean resistance with the plate pressed to the soil surface was 5,430  $\Omega$ . When the plate was rested on the leaves with no soil contact the resistance was 9.3 times higher at 50,400  $\Omega$ . This led to the current being only 13.7% and the average discharged energy being 20% of the equivalent for soil contact.

### **4.5.3     *Amaranthus powellii***

Two trials applied treatments to *A. powellii* seedlings. The first trial (Trial 4) used 4.5 kV treatments applying 50 or 100 pulses of 100  $\mu\text{s}$  duration or 100 or 200 pulses of 200  $\mu\text{s}$  duration with the flat plate electrode either on the leaves only or pressed to the soil. An additional treatment applied 10 x 1,000  $\mu\text{s}$  pulses to plants pressed to the soil. All but one of the treated plants died within 7 days, and the last outlier after 23 days. Even the weakest 4.5 kV treatment with 50 x 100  $\mu\text{s}$  pulses applied to soil killed all plants. A dose of 3 J applied to leaves only, or 5 J applied to plants pressed to dry soil, was sufficient to kill all plants. A Kruskal-Wallis test was completed with the “Life Days” of the surviving plants set to 25, the end of observations when survivors were completely healthy and growing. All treatments were significantly different to the control plants ( $H(9) = 29.89$ ,  $p < 0.001$ ). The soil-applied 4.5 kV, 100 x 100  $\mu\text{s}$  treatment which died faster was different to leaf applied 4.5 kV, 50 x 100  $\mu\text{s}$  pulse treatment which lasted longer.

The second trial (Trial 5) repeated the 4.5 kV voltage level, applying 25 or 50 x 100  $\mu\text{s}$  pulses with the flat plate electrode either on the leaves only or pressed to the soil, and 5 x

250  $\mu$ s, 500  $\mu$ s or 1000  $\mu$ s pulses pressed to the soil. An additional 3.0 kV treatment applied a single 5000  $\mu$ s pulse pressed to the soil. Some leaf treated plants and 35 of 36 soil treated plants died (Figure 4-10). The surviving soil-treated plant had received the lowest measured energy discharged (1.13 J) of any soil-treated plants, a level similar to leaf treated plants that did not die. All plants receiving more than 3 J treatments died. A logistic regression was performed to ascertain the effects of treatment on the likelihood that plants would be killed. The logistic regression model was statistically significant,  $\chi^2(5) = 27.425$ ,  $p < 0.001$ . The model explained 58.4% (Nagelkerke  $R^2$ ) of the variance in outcome and correctly classified 85.2% of the cases.

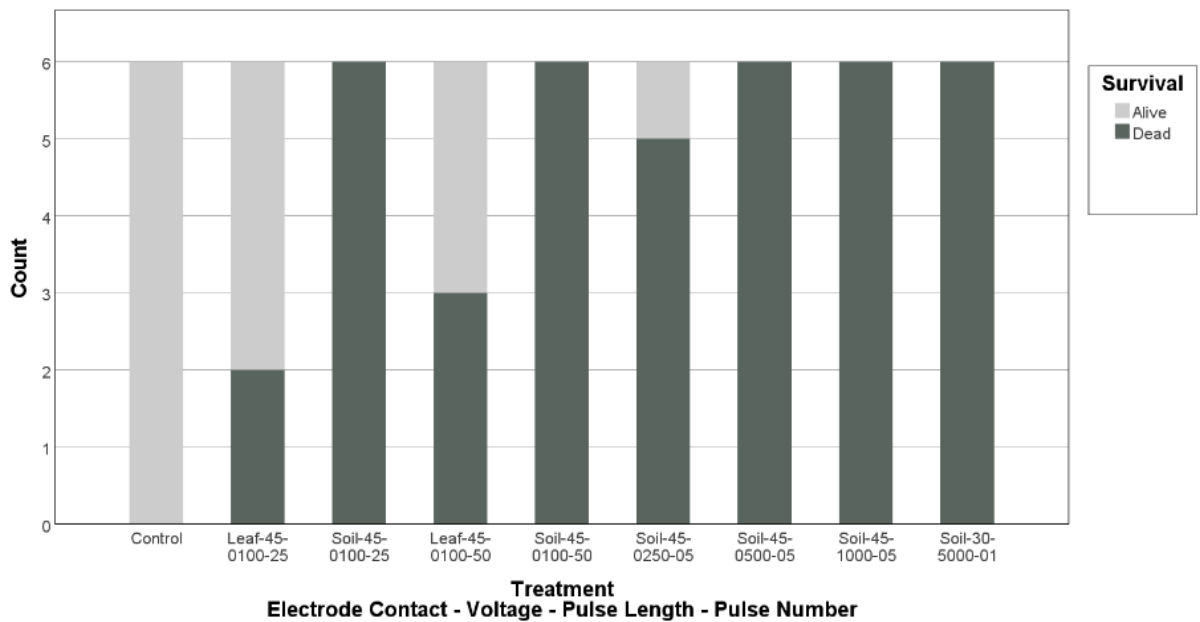


Figure 4-10 Stacked histogram count of treatment by survival of *Amaranthus powellii* following application of treatments at 4.5 kV with different electric pulse lengths and number of pulses applied with the electrode pressed to the leaves only or to the leaves pressed against the soil.

Electrical measurements showed significant differences when the same dose applied with the plate electrode pressed to the soil surface, discharging at least twice as much energy (Figure 4-11) e.g. 4.5 kV, 25 x 100  $\mu$ s pulses ( $p=0.012$ ) and 4.5 kV, 50 x 100  $\mu$ s pulses ( $p=0.032$ ).

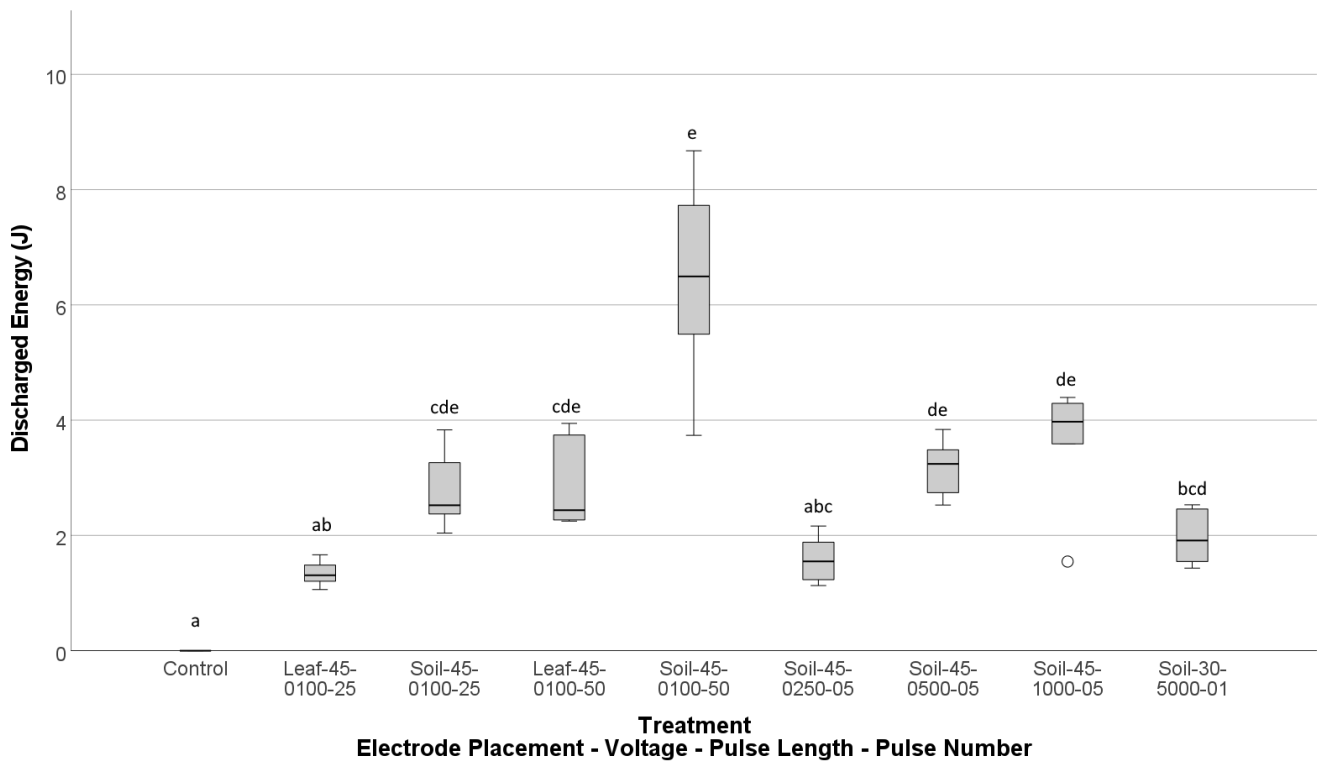


Figure 4-11 Clustered boxplot of measured energy (J) discharged when *Amaranthus powellii* were subjected to treatments at 3.0 kV and 4.5 kV with different electric pulse lengths and a different number of pulses applied to plants with the electrode pressed to the leaves only or to the leaves pressed against the soil. A mild outlier is indicated by the small circle. Plots sharing the same letters are not significantly different.

A Kruskal-Wallis test was completed with the longevity of the surviving plants set to 28 days, the end of observations when survivors were completely healthy and growing (Figure 4-12). It showed there was a significant effect of treatments ( $H(8) = 25.269, p < 0.001$ ). Unadjusted significant differences were found between the untreated control and all soil-applied treatments but not with the leaf-applied treatments. There were significant differences in longevity between the lower energy leaf treatment at 4.5 kV with 25 x 100  $\mu$ s pulses and soil treatments at 4.5 kV with 5 x 500  $\mu$ s pulses, 4.5 kV with 5 x 1000  $\mu$ s pulses, 4.5 kV with 50 x 100  $\mu$ s pulses, and 3.0 kV with 1 x 5000  $\mu$ s pulse, but not at 4.5 kV with 5 x 250  $\mu$ s pulses or 4.5 kV with 5 x 500  $\mu$ s pulses. The higher energy leaf treatment at 4.5 kV with 50 x 100  $\mu$ s pulses was significantly different to soil treatment

at 4.5 kV with 5 x 1000  $\mu$ s pulses, but not to other treatments. The more sensitive Jonckheere-Terpstra test also indicated significant difference between the control and stronger leaf treatment at 4.5 kV with 50 x 100  $\mu$ s pulses.

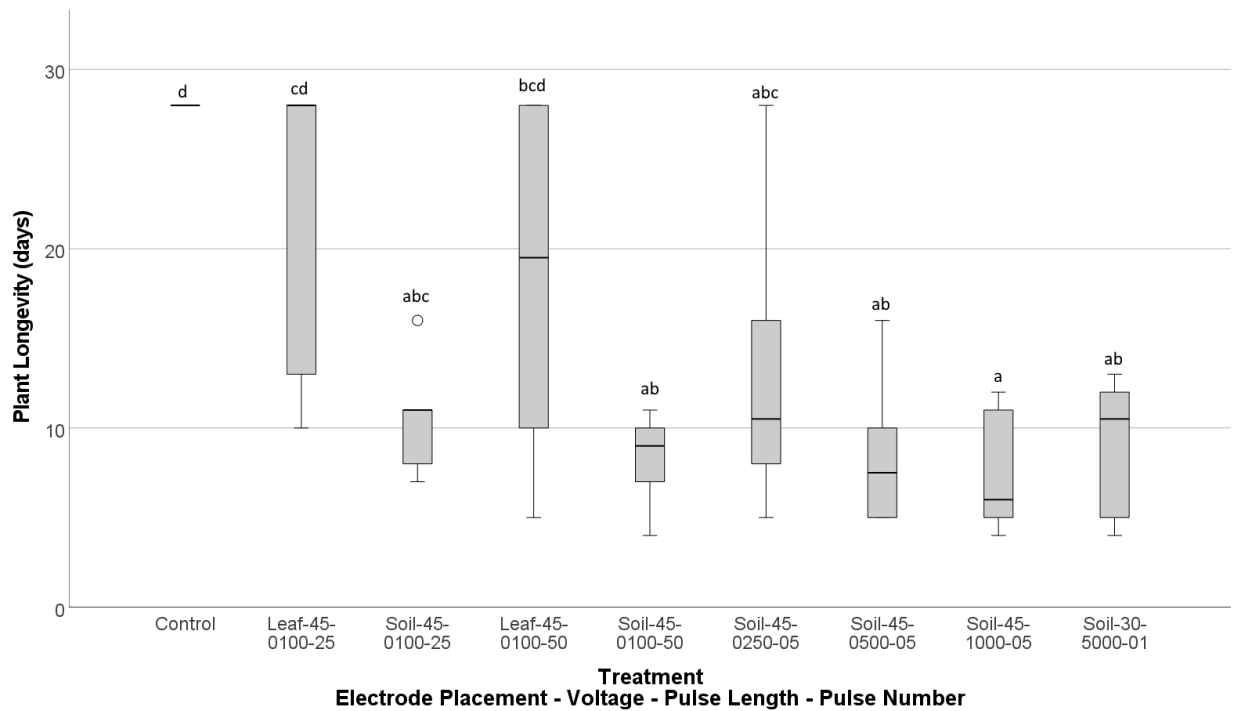


Figure 4-12 Simple boxplot of plant longevity when *Amaranthus powellii* were subjected to treatments at 3.0 kV and 4.5 kV with different electric pulse lengths and a different number of pulses applied to plants with the electrode pressed to the leaves only or to the leaves pressed against the soil. Plant assessment stopped 28 days after treatment. A mild outlier is indicated by the small circle. Plots sharing the same letters are not significantly different.

Final plant dry mass measurements included dead tissues so dead plants have a positive recorded weight, reflecting any growth at treatment plus additional biomass accumulation before death (Figure 4-13). A Kruskal-Wallis test showed significant effects of treatment ( $H(8) = 19.915, p < 0.011$ ). Unadjusted significant differences were found between the untreated control and all treatments except the lowest energy leaf applied treatment at 4.5 kV with 25 x 100  $\mu$ s pulses. The lower energy leaf treatment at 4.5 kV with 25 x 100

$\mu\text{s}$  pulses was significantly different to the soil treatments at 4.5 kV with 50 x 100  $\mu\text{s}$  pulses and 4.5 kV with 5 x 1000  $\mu\text{s}$  pulses.

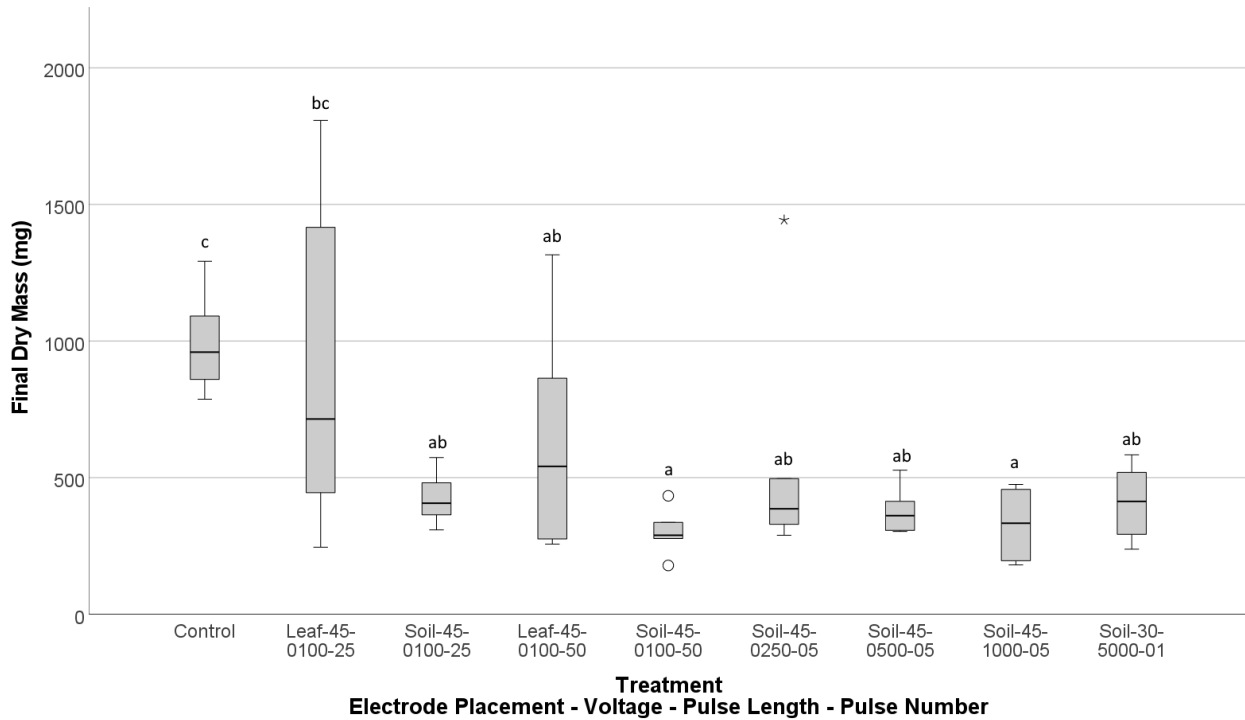


Figure 4-13 Simple boxplot of *Amaranthus powellii* final plant dry mass when plants were subjected to treatments at 3.0 kV and 4.5 kV with different electric pulse lengths and a different number of pulses applied to plants with the electrode pressed to the leaves only or to the leaves pressed against the soil. An extreme outlier is indicated by the star. Mild outliers are indicated by the small circles. Plots sharing the same letters are not significantly different.

#### 4.5.4 *Amaranthus deflexus*

The third associated trial (Trial 6) using slightly smaller *A. deflexus* applied 50 x 100  $\mu\text{s}$  pulses at 3.0 kV with the flat plate electrode pressed on to the plant and soil surface. All treated plants and no control plants died. The energy discharged averaged 1.64 J and was not significantly different to the same voltage single 5,000  $\mu\text{s}$  pulses applied to *A. powellii* in the same soil type and bags which averaged 2.46 J (ANOVA  $p=0.369$ ).

#### 4.5.5 *Solanum nitidibaccatum*

Trial 7 with *S. nitidibaccatum* applied four increasing energy treatments with the electrode pressed to leaves only and with the electrode pressing plants to the soil surface. Energy levels were based on changing voltage and pulse number. The voltages used were 4.500 kV and 3.18 kV, which in theory has half the energy. Either 25 or 50 x 100  $\mu$ s pulses were applied. A ninth treatment applied a single 5,000  $\mu$ s pulse to leaves only at 3.18 kV, theoretically the same energy as 50 x 100  $\mu$ s pulses.

When electrical doses were applied with the electrode pressed to the plant leaves only, 18 of 24 plants died. All but one of the plants treated with 4.5 kV died. Only two of 24 plants given the same doses died when the electrode pressed the plant against the soil surface. None of the plants treated with a single long pulse died. The surviving leaf-treated plant received the lowest discharge of the treatment (3.6 J compared to the 5.5 J treatment average). All plants receiving leaf contact applications with discharge greater than 3.6 J died. The energy discharges for the two soil-pressed plants that died were 9.9 J and 19.9 J.

A Kruskal-Wallis test showed significant effects of treatment on final dry mass ( $H(9) = 21.497, p = 0.011$ ). However, only two 4.5 kV leaf applied treatments, 4.5 kV with 25 x 100  $\mu$ s pulses ( $p=0.014$ ) and 4.5 kV with 50 x 100  $\mu$ s pulses ( $p=0.002$ ) were significantly different to the control. There was no significant biomass difference between the control and treatments with the electrode pressing the plant to the soil.

Higher energy doses gave higher measured energy discharges in all equivalent cases (Figure 4-14).

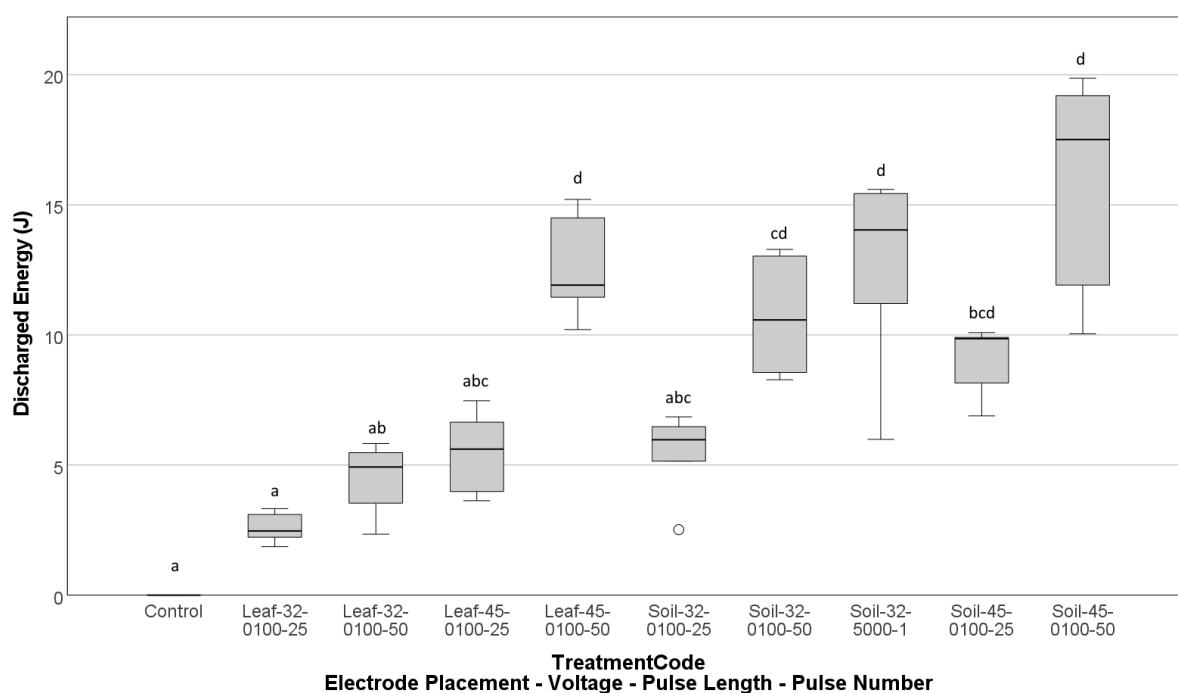


Figure 4-14 Clustered boxplot of discharged energy (J) by dose applied by electrode placement when PMS was applied to *Solanum nitidibaccatum*. A mild outlier is indicated by the small circle. Plots sharing the same letters are not significantly different.

At 3.18 kV the treatments applied with plants pressed to the soil discharged 226% more energy than the same dose applied to the plant leaves only. At 4.5 kV the soil discharges were 145% higher than leaf discharges. Only one pairing of treatments that were expected to have the same discharge, leaf-applied and soil-applied 3.18 kV with 50 x 100  $\mu$ s pulse doses, showed the possibility of a significant difference when analysed independently (Sig. = 0.017), but when the Bonferroni correction was applied, the difference was insignificant (Adj. Sig. = 0.776) (Table 4-2). The leaf-applied 4.5 kV with 50 x 100  $\mu$ s pulses treatment killed all plants with an average energy discharge of 12.5 J plant<sup>-1</sup>, equating to 0.625 MJ ha<sup>-1</sup> at five plants m<sup>-2</sup>. The energy discharge of energy equivalent pairs with different voltage balanced by the number of pulses, 3.18 kV with 5 x 100  $\mu$ s pulses and 4.5 kV with 25 x 100  $\mu$ s pulse doses, were 21.9% higher and 14.9% lower in the cases of leaf and soil applications, respectively.

Table 4-2 Comparison of selected electrical dose applications to *Solanum nitidibaccatum* showing mean discharged energy, difference between treatment pairs and Kruskal–Wallis pairwise comparisons of selected treatments with (Adj.Sig.) and without Bonferroni corrections (Sig.).

Treatment		Mean Value			Pairwise Comparisons of Treatments*				
Treatment 1	Treatment 2	Treat 1	Treat 2	Difference	Test Statistic	Std. Error	Std. Test Statistic	Sig.	Adj. Sig. <sup>a</sup>
Leaf-32-0100-25	Soil-32-0100-25	2.573	5.491	2.134	-12.333	10.078	-1.224	0.221	1.000
Leaf-32-0100-50	Soil-32-0100-50	4.505	10.720	2.380	-24.000	10.078	-2.381	0.017	0.776
Leaf-45-0100-25	Soil-45-0100-25	5.493	9.127	1.662	-12.667	10.078	-1.257	0.209	1.000
Leaf-45-0100-50	Soil-45-0100-50	12.535	16.010	1.277	-5.500	10.078	-0.546	0.585	1.000
Leaf-32-0100-50	Leaf-45-0100-25	4.505	5.493	1.219	4.505	5.493	1.219	0.620	1.000
Soil-32-0100-50	Soil-45-0100-25	10.720	9.127	0.851	10.720	9.127	0.851	0.530	1.000
Soil-32-0100-50	Soil-32-5000-01	10.720	12.717	1.186	-5.167	10.078	-0.513	0.608	1.000
Soil-45-0100-25	Soil-32-5000-01	9.127	12.717	1.393	11.500	10.078	1.141	0.254	1.000

\* Each row tests the null hypothesis that the Sample 1 and Sample 2 distributions are the same. Asymptotic significances (2-sided tests) are displayed. The significance level is .050.  
a. Significance values have been adjusted by the Bonferroni correction for multiple tests.

## 4.6 Discussion

The objectives of this study were to determine whether a flat plate used in a practical, ‘paddock-treatment’ way could apply a threshold dose of voltage and energy to weed seedlings to achieve > 90% mortality, to assess energy expenditure, and to compare relative responses of different species. Two electrode placements were tested: the plate pressed against the plant only with a 3 cm separation from the soil surface, and the plate pressing the plant on to the soil, with the soil surface either wet or dry. As in my earlier trials (Bloomer et al., 2022), I found different species had different sensitivities to PMS, a result similar to the findings of Lati et al. (2021) who tested seedlings of analogous species: *Lolium rigidum*, *Sorghum halepense*, *Amaranthus retroflexus*, and *Portulaca oleracea*.

The flat plate method failed to adequately control tillering *L. multiflorum* plants with the best treatment killing only half the treated plants, much less than the target rate of 90% control. However, the flat plate treatment with PMS had a limiting effect on growth and significantly reduced growth rates. A linear trend line with the individual 90% confidence lines fitted to trial data suggests complete control of *L. multiflorum* can be achieved at about 200 J or 10 MJ ha<sup>-1</sup>. While the trial treated three plants with each application, this does not imply that the per-plant dose may be 70 J, because much of the energy is directly lost to the soil. However, if successfully treating an average of three plants per application was achieved, the energy requirement would be 3.5 MJ ha<sup>-1</sup> if treating five plants m<sup>-2</sup>.

While monitoring the plants post-treatment, I observed that if any tiller emerged, the plant recovered. However, the subsequent growth delay may allow crop plants growing alongside treated grass weeds to develop unimpeded. I previously suggested that

insufficient energy reached the meristems of *L. multiflorum* at or below ground level within the stem sheath, enabling newly developing tillers to survive (Bloomer et al., 2022). This was also postulated by Lati et al. (2021) who reported an 85% biomass reduction when *L. rigidum* plants were treated 2 weeks after sowing. They do not provide information on plant size at treatment or whether any plants died. My earlier work treated younger *L. multiflorum* plants that in most cases were not tillering and found 3 kV was more effective than 6 kV when a point electrode was applied to leaves (Bloomer et al. 2022). The trials reported here used 3.0, 3.5 and 4.5 kV treatments and did not show conclusive evidence that voltage is a key determinant of plant death rate or post-treatment biomass.

Treating broadleaf weed seedlings was effective, and all trials showed PMS could achieve the target > 90% kill at < 2 MJ ha<sup>-1</sup>. Different broadleaf species of approximately similar size required different doses to achieve the target kill-rate with, in decreasing energy order, *P. aviculare* > *S. nitidibaccatum* > *A. powellii* > *A. deflexus*. A single leaf-applied treatment killed all *P. aviculare* weeds with 17.9 J plant<sup>-1</sup>, equivalent to 0.895 MJ ha<sup>-1</sup> if treating five plants m<sup>-2</sup>. While not tested, data for soil-applied PMS suggest the 90% target could be achieved by increasing the pulse length to 200 µs, the same as the leaf treatment, but the energy discharge would be far greater.

Treatments applied only to leaves of *S. nitidibaccatum* were more effective than the same doses applied to leaves pressed to the soil. The most effective treatment was leaf-applied and killed all plants, with an average energy discharge of 12.5 J plant<sup>-1</sup> equating to 0.625 MJ ha<sup>-1</sup>. When treated with half the dose, 83% of plants were killed. Only one plant died when these doses were applied with the electrode pressing the plant to the soil. My

earlier point-electrode trials (Bloomer et al., 2022) showed PMS to be less effective when treating the related species *S. nigrum* with no treatment killing all plants. These are fleshy species with ability to regenerate roots from aerial stem tissues. I observed that plants that successfully established new roots into the soil were able to recover, although their growth was delayed. Similarly, Lati et al. (2021) noted biomass reduction of the fleshy *Portulaca oleracea* but did not report deaths.

Treatments applying only 3 J to *A. powellii* plants by leaf contact only, or 5 J to plants pressed to the soil, were fully effective in Trial 4 with a kill rate of 100%. In Trial 5, treatments applied to plants pressed to the soil killed all plants with an average 2.76 J plant<sup>-1</sup>, equivalent to 0.138 MJ ha<sup>-1</sup>. The same applications to leaves only discharged 1.34 J plant<sup>-1</sup>. All plants receiving more than 3 J died. Although slightly smaller when treated, the closely related *A. deflexus* seedlings all died when they received a treatment with the flat plate electrode pressed on to the plant and soil surface and discharged energy of an average of 1.64 J plant<sup>-1</sup>, equivalent to 0.082 MJ ha<sup>-1</sup>.

Electrical weeding technology typically uses very high electrical current treatments and attributes control to resistive heating causing cell damage, boiling, and disruption (Diprose & Benson, 1984; Diprose et al., 1985; Vigneault & Benoît, 2001). These methods are characterised by rapid impact on plants with steam or flames often reported. My very low-energy treatments do not exhibit the same instantaneous effects, often taking several days or more for symptoms to become apparent. While I have not determined a mechanism for death, there is no obvious evidence of cellular boiling, and the applied energy seems inadequate to severely raise tissue temperatures. Lehnhoff et al. (2022) note that ‘mechanisms by which plants die by an electric current are not well understood’.

Their work applied relatively low currents to large plants and trees for a very long duration and effected control. This leads me to ponder if some other plant response is triggered by the treatments, eventually leading to plant death.

I note that my use of a flat plate electrode will generate an electrostatic field between the plate and the ground, which may affect the plant's cells and membranes (Diprose et al., 1984; Xie et al., 2000; Costanzo, 2008; Schmiedchen et al., 2018; England & Robert, 2022). Application of an external electric field can control the elongation of shoots and roots (Hamada et al., 1992) and even sub-lethal electric currents can act as abiotic elicitors that increase the production of secondary plant metabolites (Kaimoyo et al., 2008) as for a disease response. The role of secondary metabolites in programmed cell death is variously reported (Gill & Tuteja, 2010; Vartapetian et al., 2011; Petrov et al., 2015) suggesting this is an area where further research is warranted.

My goal is a weeding system that can selectively remove weeds from field crops without negative impact on crop plants. My flat plate electrode is narrower than vegetable and arable crop row-spacings and could be drawn between them and in a robotic deployment, it could apply PMS only where weeds are present. I did not find evidence of significant damage to soil microbiota in the literature dealing with high-energy electric weeding, and I postulate that my very low-energy system is unlikely to have significant adverse effects. I have observed earthworms coming to the surface and moving away from treatment sites when I have applied repeated doses in my trial work. The flat plate electrode system would be less suited to intra-row weeding of very high-population crops that have close interplant spacings. I have yet to test for negative effects on adjacent crop plants but note that the much higher-powered systems for vineyard and orchard strip-weeding by Zasso

Corporation (2023) and others have shown no impacts on adjacent plants and leave a well-defined edge between treated and untreated sward zones.

## 4.7 Conclusions

These experiments used a flat-plate electrode to apply PMS to a range of species, using different voltages, pulse lengths, and pulse numbers to plants grown in bags in a greenhouse. They further demonstrate the potential of PMS as an ultra-low-energy treatment method for controlling small non-tillering grasses and a range of broadleaf weed species at the seedling stage. Very short, high-voltage DC treatments require far less energy than alternative weed control options, including currently available electrical weeding systems, and I achieved better control using less time and energy than other low-energy electrical weed treatment methods that have been reported. In these trials, my method killed only half of the treated *L. multiflorum* “Winter Star” plants, well below my target rate, but it significantly reduced growth rates and indicated that an effective treatment of  $< 2.0 \text{ MJ ha}^{-1}$  for treating five plants  $\text{m}^{-2}$  may be possible. *Polygonum aviculare*, *A. powellii*, *A. deflexus*, and *S. nitidibaccatum* plants were successfully controlled using from 0.1 to 0.9  $\text{MJ ha}^{-1}$ , indicating that broadleaf weeds are more susceptible than grasses, although different species require different doses. This result easily met my target effectiveness and efficiency goals. Although broadleaf weeds appear easier to kill than grasses, fleshy broadleaf species with the ability to produce aerial roots may be more difficult to control. Treatment is more energy-efficient when the electrode is separated from direct ground contact. The application with the plate electrode pressing on a wet soil surface used far more energy than that on a dry soil surface. Given the overall extremely low energy use, there is a practical opportunity for increasing doses to effect better seedling kill rates.

While these trials were conducted in a laboratory setting, with precise field application, the results should be replicable, and this is the subject of my ongoing research (Chapter 5). The ability to apply PMS using a flat-plate electrode pressed to the plant leaves or plants and soil removes the issue of the very precise targeting required when using a point electrode. Coupled with the small energy requirement, it makes the system suitable for incorporation in a mobile hand-held weeding tool or individually or in groups in a precision robotic system to control herbicide-resistant weed seedlings that have escaped chemical or other control methods. Voltages have been kept below 10 kV, and the energy released by the equipment is low. In general, these would place the equipment used within the criteria acceptable for electric fence energisers (International Electrotechnical Commission, 2002; Gallagher Group, 2018).



# Chapter 5. Field Deployment

MICROSHOCK WEED CONTROL IS EFFECTIVE AND ENERGY EFFICIENT IN THE FIELD.

## 5.1 Publication

This chapter was published as:

Bloomer, D. J., Harrington, K. C., Ghanizadeh, H., & James, T. K. (2024a). Pots to Plots: Microshock Weed Control Is an Effective and Energy Efficient Option in the Field. *Sustainability*, 16(11), 4324. (Bloomer et al., 2024)

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Flat plate weeding is an effective and ultra-low energy weed control suitable for manual, robotic, or conventional deployment without recourse to tillage or chemical herbicides. It gave better than 90% control with energy to treat 5 weeds m<sup>-2</sup> equivalent to 25 kJ ha<sup>-1</sup> for *L. didymum* and *A. powellii*, and 555 kJ ha<sup>-1</sup> for in-ground *L. multiflorum*, all well below my 1 MJ ha<sup>-1</sup> target and a fraction of the energy required by any other weeding system. This is the first study to compare applications to the leaves only or to leaves pressed against the soil surface, to seedlings growing outside in the ground and to plants growing in bags of the same soil, and showed that greenhouse and in-field results are comparable, other factors remaining constant.

**Statements of Contribution:** See Appendix 1 Statements of Contribution

## 5.2 Abstract

Seeking low environmental impact alternatives to chemical herbicides that can be integrated into a regenerative agriculture system, I developed and tested flat plate electrode weeding equipment applying ultra-low energy electric shocks to seedlings in the field. Better than 90% control was achieved for all species, with energy to treat 5 weeds m<sup>-2</sup> equivalent to 15 kJ ha<sup>-1</sup> for *L. didymum* and *A. powellii*, and 363 kJ ha<sup>-1</sup> (leaf contact only) and 555 kJ ha<sup>-1</sup> (plants pressed to soil) for in-ground *L. multiflorum*, all well below my 1 MJ ha<sup>-1</sup> target and fraction of the energy required by any other weeding system. I compared applications to the leaves only or to leaves pressed against the soil surface, to seedlings growing outside in the ground and to plants growing in bags filled with the same soil. No previous studies have made such direct comparisons. My research indicated that greenhouse and in-field results are comparable, other factors remaining constant. The in-ground, outdoor treatments were as effective and efficient as my previously published in-bag, greenhouse trials. The flat plate system tested supports sustainable farming by providing ultra-low energy weed control suitable for manual, robotic, or conventional deployment without recourse to tillage or chemical herbicides.

**Keywords:** electric weeding; sustainable farming; non-chemical weed control; electrodes; robotic weeding; discharge energy; weeds.

### 5.3 Introduction

Sustainable farming and regenerative agriculture (LaCanne & Lundgren, 2018; Burns, 2020; Grelet & Lang, 2021; O'Donoghue et al., 2022; Rempelos et al., 2023) are frameworks of principles and practices that address critical issues such as degraded soils and reduced water quality (Hageman et al., 2019), pest resistance, and dependence of non-renewable energy supplies (Jayasinghe et al., 2023). Commenting in 1995 on weed science in sustainable agriculture, Zimdahl (1995) questioned agricultural practices that “largely ignored ecological concerns” and that addressed problems through increasing applications of fertilisers, pesticides, water and energy, and suggested a need to move from reliance on chemical technology. The flow of publications of regenerative agriculture has continued and more recent work has sought to define and validate regenerative farm systems (Fenster et al., 2021) and investigate opportunities and constraints of certification (Elrick et al., 2022), and to introduce industry certification schemes (McCain Foods, 2023). Common among these is recognition of the need to reduce use of chemical pesticides including herbicides and to minimise tillage. The potential impact of agrichemicals on workers, consumers and the environment can be compared using the environmental impact quotient (EIQ) (Cornell University College of Agriculture and Life Sciences, 2024). In a separate LandWISE research project, we have calculated the EIQ for process tomatoes crops grown in New Zealand and found that herbicides accounted for about 1/3<sup>rd</sup> of the overall score. The need for alternative, non-chemical weed control methods has been well described in Chapter 2 (Bloomer et al., 2023b), with consumer preference (Forbes et al., 2009; Crinnion, 2010; Hoek et al., 2017; Koch et al., 2017; Galati et al., 2019), legislative restrictions and public concern about environmental effects (Bajwa et al., 2015; Jun et al., 2023), development of regenerative growing systems (Ministry for Primary Industries, 2022) and herbicide resistance (Heap,

2020; Buddenhagen et al., 2021; Ngow, 2022; Doole & James, 2023; Espig & Henwood, 2023) noted as drivers. I also note the need to reduce soil tillage and energy consumption (Alluvione et al., 2011), and particularly to reduce dependence on fossil fuels.

As a weed scientist, I have been seeking ultra-low energy, non-herbicide weed control methods that can be implemented within a regenerative farming system. Dismissing tillage and high energy systems such as flaming, steaming and laser treatment, I have focused on very low energy electrical weeding techniques. I have shown that electric weeding with pulsed electric microshocks (PMS) is an effective ultra-low energy option that requires a fraction of the energy of any other system (Bloomer et al., 2022). I demonstrated in greenhouse trials using a precisely placed point-electrode that a minimal amount of electrical energy was sufficient to control small, non-tillering grasses and broadleaf weed seedlings up to 15 cm in height. At 5 J plant<sup>-1</sup>, a density considered reasonable for escape weeds surviving after a chemical herbicide or mechanical treatment (Kaufman & Schaffner, 1982; Vigneault et al., 1990; Coleman et al., 2019), my system treating five plants m<sup>-2</sup> would require 0.25 MJ ha<sup>-1</sup> plus transport and actuation energy. Seeking an easy-to-deploy method, I then tested a flat-plate electrode to apply PMS to a grass and four broadleaf weed species (Bloomer et al., 2023a). I found that tillering *Lolium multiflorum* Lam. (Italian ryegrass) was more difficult to control, but a range of broadleaf weeds were successfully managed, with the energy required to kill 100% of seedlings varying from 0.1 to 0.9 MJ ha<sup>-1</sup>. This was still a very low energy requirement compared to other weed control options (Kaufman & Schaffner, 1982; Vigneault et al., 1990; Fergedal, 1994; Goell et al., 2003; Helsen & Pimentel, 2007; Alluvione et al., 2011; De Cauwer et al., 2015; Gonzalez-de-Soto et al., 2015; Coleman et al., 2019; Andreasen

et al., 2021; Coleman et al., 2021; Lati et al., 2021; Denmukhammadiev et al., 2022; Mwitita et al., 2022).

There is little information comparing treatment of plants with electricity in laboratory or greenhouse settings with treatment of similar plants in outdoor field settings. Diprose et al. (1978) suggested that maybe two to three times more power was required to kill plants in the field compared to those grown in pots in a greenhouse, disagreeing with Chandler (1978) who suggested that ten times more energy is required to kill plants in the field relative to those indoors. However, in each case the plants being compared were not equal. Chandler noted considerable variation based on species and age, and Diprose et al. proposed the difference they saw was due to plant size. Other electrical weeding research typically describes plant size, but not necessarily in comparable ways, making it difficult to compare different results. While some give physical measurements such as stem diameter and plant height (Mizuno et al., 1990), or number of leaves (Blasco et al., 2002), plant age has also been used as a size descriptor. For example, Lati et al. (2021) described pot experiments with weeds at various numbers of weeks after seeding. No physical size measurements at time of treatment were reported, although final biomass measurements were given. The size of contact area of the root surface with the growing medium was also found to be related to overall electrical resistance (Cao et al., 2010) and hence the energy needed to apply a certain dose to plants.

Electrocution of in-ground plants has been theorised since the 1890s (Sharp, 1893; Scheible, 1895) and commercially available since the 1970s (Vigoureux, 1981; Kaufman & Schaffner, 1982; Diprose & Benson, 1984; Diprose et al., 1985). However these are all very high energy systems. Blasco et al. (2002) investigating robotic weed control system

architecture, treated weeds in a field crop with 15 kV shocks at 30 mA for 200 ms (90 J) and eliminated all weeds with fewer than five leaves or less than 20 cm tall and “no significant damage was caused to lettuces having more than ten leaves”. They did not report equivalent indoor or in-bag treatments. Mizuno et al. (1990) developed an apparatus for laboratory application and showed small plants (40-60 mm height, 1-3 mm stem diameter) could be destroyed by one spark discharge of 0.14 J energy and very large plants (800-1,200 mm height, 10-15 mm stem diameter) with repeated pulses totalling only 2 J. They also investigated use outdoors, describing a low-energy spot-weeder powered by a 12 V battery to kill *Poa annua* L. (annual poa) in golf courses using a 3 kV 200 W high frequency alternating current (AC) discharge (Mizuno et al., 1993). However the different experiments are not linked to compare *in vitro* with *in terra* effects.

I have previously applied approximately 6 J single pulse direct current (DC) shocks to in-ground weeds using point electrodes as part of prototype equipment testing. In that previously unreported work (see Chapter 6.4.2), treated species including *Capsella bursa-pastoris* L. (shepherd’s purse), *Sonchus oleraceus* L. (sow thistle), *Lepidium didymum* L. (twin cress) and *P. annua* showed no symptoms for about six days, after which most broadleaf plants senesced and died, but the grasses appeared harder to kill. I then completed numerous greenhouse studies using my multiple-pulse electric weeding system (Bloomer et al., 2022, 2023a).

Seeking to determine that treatment effects on bagged plants growing in a greenhouse would transfer to commercial field conditions using regenerative practices, I took my ultra-low energy PMS research outdoors and compared the effects of treatments applied to weeds grown in bags with those applied to weeds grown directly in the ground. In an

extension of my earlier studies, my main objectives were to determine whether a flat plate electrode used on outdoor, in-ground plants (field treatment) could apply a threshold “dose” of voltage and energy to achieve more than 90% mortality, to assess energy expenditure, and to compare relative responses of different species. I set a goal of attaining the energy efficiencies of my greenhouse trials; to achieve control of five weeds m<sup>-2</sup> at less than 1 MJ ha<sup>-1</sup> plus transport energy. The trials reported here included transplanted *L. didymum* and *Amaranthus powellii* seedlings, and *L. multiflorum* cv. “Winter Star” grown from directly sown seed. Comparisons of energy discharges and weed killing effectiveness are presented.

By necessity, the earthing electrode is very close to the treatment electrode and plant when trialling electric weeding on greenhouse grown bagged plants. In the field, there is no such limitation, and it may be advantageous to allow a larger distance between them to facilitate simpler machinery design and easier operation. Therefore, the effect of increasing electrode distance was tested. Similarly, it may be easier to deploy a surface-contact earthing-electrode rather than a soil-inserted one which may become clogged with debris, so a flat disc and a probe were compared.

## **5.4 Materials and methods**

### **5.4.1 Equipment and Materials**

A custom-built PMS device that produces multiple direct current (DC) pulses of up to 4.5 kV was developed by Weda Tech Ltd. (Hastings, New Zealand) (Figure 5-1A).

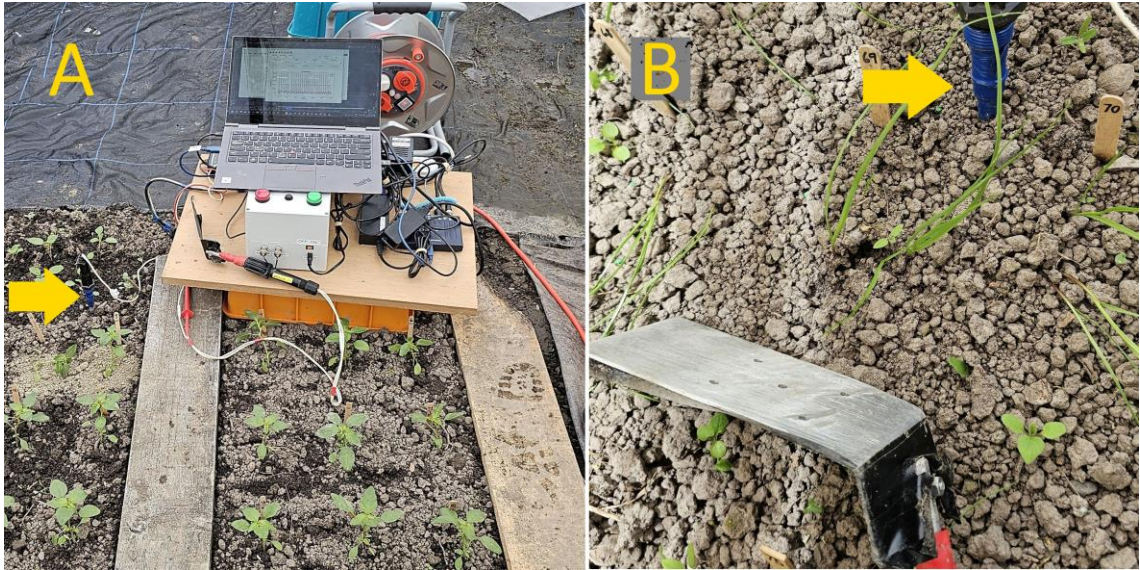


Figure 5-1 Photographs showing the WedaTech PMS equipment set up ready to apply flat plate electrode treatments to (A) in-ground grown *Amaranthus powellii* seedlings, and (B) sets of three *Lolium multiflorum* seedlings as single applications. The earthing electrode (yellow arrow) is the 5 mm diameter rod inserted 75 mm into the soil.

The device was controlled using custom software running on a laptop, with the discharge voltage, pulse duration, pulse period and number of pulses able to be programmed. This sets the potential level of applied energy, but variations in the resistances of plants and soil affect discharge current so the actual energy discharged varies. To ensure full capacitor recharge, a 50 ms interval was maintained between pulses. A PicoScope 2000 series oscilloscope was coupled with a Pico TA044 high-voltage differential probe connected to the positive and earthing electrodes to monitor the applied doses. Pulse data were automatically recorded and logged in comma separated value (csv) files.

The first trial compared treatments applied to bagged *L. didymum* plants growing in a greenhouse with treatments applied to in-ground plants growing in a field. It also compared two earthing electrode types, having the earthing and application electrodes in different locations along a transect, and having the system with and without a plant as part of the circuit. The second and third trials compared the effectiveness of equivalent

treatments on outdoor bag grown *A. powellii* and *L. multiflorum* plants with plants growing in the ground beside them, and treatments with the application electrode in contact with the leaves only or with the plant pressed against the soil surface.

For the *A. powellii* and *L. multiflorum* trials, the positive application electrode was a flat rectangular aluminium plate with dimensions of 75 mm x 100 mm (Figure 5-1 B). Earthing was achieved by a 5 mm-diameter aluminium rod inserted 75 mm into the ground. For the *L. didymum* trial the positive application electrode was a 40 mm diameter aluminium disc contacting the centre of seedlings (Figure 5-2). For part of the *L. didymum* trial, an alternative earthing arrangement was also tested, replacing the inserted 5 mm rod earthing electrode (Figure 5-2 A) with a 30 mm aluminium disc pressed against the soil surface a few centimetres apart from the positive application electrode disc (Figure 5-2 B).

In all trials, the growing medium was Hastings silt loam soil, a Typic Orthic Gley (Manaaki Whenua - Landcare Research, 2024). The soil in the ground was dug manually, crumbled, and raked to leave a level but slightly rubbly surface. Soil excavated from the same area was sieved to 5 mm and used to grow the bagged plants.

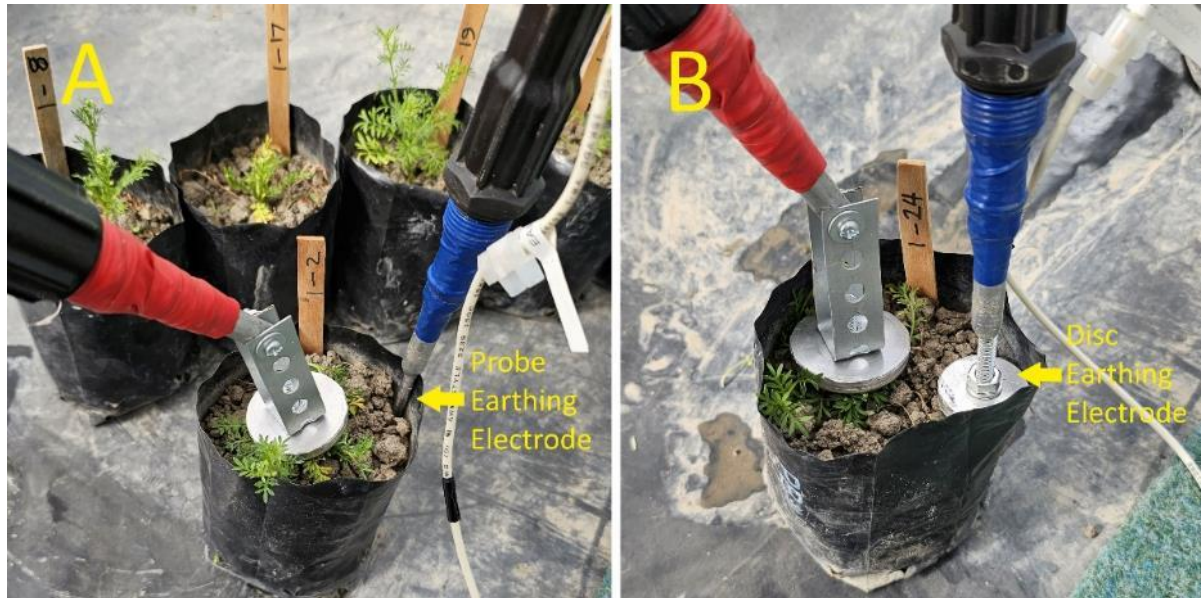


Figure 5-2 Photographs of alternative treatments being applied to bag grown *Lepidium didymum* seedlings using a 40 mm treatment electrode and (A) a 5 mm earthing probe inserted 75 mm into the soil, and (B) a 30 mm diameter disc earthing electrode pressed to the soil surface.

After treatments were applied, the *L. didymum* bagged plants were kept in a greenhouse with clear 6 mm polycarbonate panes (Winter Gardenz, Auckland, NZ). The in-ground plants were planted nearby and covered by 1 cm mesh bird netting to avoid damage by rabbits or native pūkeko birds. The *A. powellii* and *L. multiflorum* plants were grown in a temporary 3 m wide tunnel covered by 0.58 mm Cropsafe insect mesh (Cosio Industries, Auckland, NZ), with the bags placed adjacent to the in-ground plants.

#### 5.4.2 Environmental data

Temperature, wind, radiation, rain, and humidity were logged by the Ruahapia weather station (HortPlus Ltd., Hastings, New Zealand) sited 100 m away from the plots, and showed no values considered likely to cause any negative effects.

Soil moisture percentage for in-ground grown plants at time of treatment was determined by multiple readings taken using a 200 mm probe-length Hydrosense II sensor (Campbell

Scientific Inc., Logan, Utah, USA) inserted into the soil in the vicinity of the plant and earthing electrode positions. For bag-grown plants, the bags were weighed at time of treatment. At the end of the trial, the soil was oven dried at 35 °C for three days, and percentage moisture determined from the difference. Noting in the *L. didymum* trial that dry soil acted as electrical insulation, the subsequent *A. powellii* and *L. multiflorum* trials had a thin layer of approximately 5 mm of dry soil added to the wet soil surface before the soil-pressed treatments were applied, with the aim to minimise direct loss of applied electricity to the soil body. The added soil was included when determining dry weight at the end of the trial.

When *L. didymum* plants were treated, the in-bag soil moisture was uniform with a mean of 34.6% (n = 14, SD = 0.03). The in-ground soil had high variability in moisture content with depth and while the soil surface had dried peds, the underneath was damp and very moist at depths below about 100 mm. The mean moisture in the top 200 mm was 36.2% (n = 10, SD = 1.2%). At treatment time, the soil moisture in bagged *A. powellii* plants was 32.7% (n = 84, SD = 3.2), slightly higher (p = 0.04) than in ground-grown plants at 31.3% (n = 28, SD = 2.9). Rain in the days prior to treatment caused soil moisture differences between bagged and in-ground *L. multiflorum* plants. Bagged plants did not drain well and had a significantly higher (p < 0.001) mean moisture content of 41.7% (n = 73, SD = 5.0) than in-ground plants with a mean soil moisture content of 27.1% (n = 47, SD = 4.7). There was no significant difference in soil moisture between plants within the in-ground treatments, nor between plants within the bagged treatments.

### 5.4.3 Methods

For each application, the pulse voltage and current were recorded and the discharged energy was calculated. Subsequent plant health was scored, and death rates were determined. Results were compared for bag versus ground grown plants, for electrode application to leaves only versus to plants pressed to the soil surface, and by applied dose.

Trial 1 compared treatments applied to *L. didymum* seedlings with four to eight leaves taken from a cropping field and grown individually in 450 mL bags in a greenhouse or in the ground outdoors at 150 x 200 mm spacing. At 7 days after treatment (DAT), *L. didymum* plants were scored for vigour with a score of 1 accorded to fully healthy plants and a score of 0 to dead plants. Plant vigour assessment was subjective based on leaf colour, turgor and rosette growth with equal weightings. At 23 DAT final assessments were made including survival (dead or alive), days to death, stem state, leaf and side shoot colour and turgidity. All the plants that were not obviously dead were classified as alive.

The effect of the soil itself on the amount of energy discharged by the machine was tested during the outdoor *L. didymum* trial, firstly by pairing earthing and positive treatment electrode rods (inserted 10 cm apart and 75 mm into the soil) at different locations along a transect, and secondly by inserting the earthing electrode probe 75 mm into the soil and applying the positive treatment electrode at increasing distances away (Figure 5-3). To test the influence of earthing electrode types with or without plants as part of the circuit, two options (a 5 mm diameter rod inserted 75 mm into the soil and a surface-pressed 30 mm diameter disc) the process was repeated with the disc treatment electrode pressing plants to the soil surface and with no plants present.



Figure 5-3 Photograph showing (A) the WedaTech PMS equipment, and (B) an array of 5 mm aluminium probe electrodes at 10 cm spacings set up to test soil conductivity and resistance in different positions and with increasing electrode separation gaps. The positive electrode (C) is coloured red, and the negative earthing electrode (D) is coloured blue.

The resulting data were standardised by adjusting for the separation distance between the electrodes to give a value of energy per distance, expressed as  $\text{J m}^{-1}$ . Fit was determined using power function trendlines. Comparisons were also made between two earthing electrode types (a 5 mm diameter rod inserted 75 mm into the soil and a surface-pressed 30 mm diameter disc), having the earthing and application electrodes in different locations along a transect (Figure 5-3), and having the system with and without a plant as part of the circuit.

In Trial 2, *A. powellii* plants were germinated in trays in a greenhouse from field soil with a known weed infestation and transplanted individually into either 450 mL soil-filled bags or directly into the ground at 150 x 200 mm spacings. Plant status was assessed using a condition score based on subjective observations of plant vigour (50%), stem tip angle (5%), stem collapse (10%), stem diameter (10%), shoot colour (5%), shoot turgor (5%),

leaf colour (5%) and leaf turgor (10%) combined to a total possible score of 20 for fully healthy plants. A dead plant scored 0 regardless of any other ratings. Final assessments were made 17 DAT, by which time plants were clearly alive or dead.

In Trial 3, *L. multiflorum* plants were sown in sets of six, in 450 mL soil-filled bags, and in the ground with sets at 150 x 200 mm spacings. Prior to treatment, each set was thinned to three plants such that all were of similar size. The plants were measured and bagged sets of three plants were sorted into equivalent groups for treatment. Plants were visually assessed prior to treatment, with the number of green leaves and tillers counted, and the length of the longest leaf of each plant measured. Most plants had two or three leaves and one tiller, but some were starting to produce second tillers. Measurements of living plant tissues were repeated, and plant deaths were recorded until the trial was stopped 14 DAT. Severely shrivelled or dead parts of the leaf were discounted, and any totally brown and shrivelled plants were classified as dead.

#### **5.4.4 Treatments**

The doses selected for each trial were based on results from earlier greenhouse work with the same or similar species of similar size. Seeking to determine an effective dose rate, the lowest energy applied dose was expected to have little effect, and the highest energy dose was expected to kill all plants. For *L. didymum*, a range of treatments pressing the leaves to the soil surface was applied at 4.5 kV with the bagged plants receiving a dose of either 25, 50 or 100 x 100  $\mu$ s pulses. The in-ground plants were all treated with 100 x 100  $\mu$ s pulses, but with two alternative earthing arrangements employed.

The *A. powellii* plants received a 4.5 kV dose of either 25 x 25  $\mu$ s, 50 x 50  $\mu$ s, 50 x 100  $\mu$ s, 100 x 100  $\mu$ s, or 100 x 200  $\mu$ s pulses. The same treatments were applied to both bagged and in-ground plants, with the treatment electrode either on the leaves only or pressing the plant against the soil surface. A thin insulating layer of about 5 mm of dried sieved soil was applied to the moist soil surface before plants were pressed to the soil for treatment.

In my early *L. multiflorum* research (Chapter 3) I found that higher voltages could damage fine grass leaves but fail to kill growing points at ground level (Bloomer et al., 2022). In this trial, treatments were applied at either 3.5 or 4.5 kV, each with 100 x 200  $\mu$ s, 200 x 200  $\mu$ s, or 200 x 400  $\mu$ s pulses. As with *A. powellii*, the same treatments were applied to both bagged and in-ground plants, with the electrode either on the leaves only or pressing the whole plant against the soil surface which if moist, had a thin insulating layer of about 5 mm of dried sieved soil applied to the surface before plants pressed to the soil were treated. The mean plant size and treatments applied in each of the trials are summarised in Table 5-1.

#### **5.4.5 Statistical analysis**

Data were statistically analysed using SPSS Version 28.0.1.0 (142). Where differences are reported, the default for statistical comparisons of groups was the independent samples Kruskal-Wallis one-way ANOVA (k samples) because almost all cases failed the Levene's test for ANOVA homogeneity of variance. In each trial, plants were randomised, and each treatment had six replicates, a sample size satisfactory for Kruskal-Wallis (Field, 2013). For *L. didymum* and *A. powellii*, each replicate was one plant.

Table 5-1 Trial summary table showing trial number, weed species, plant size, and treatments applied to *Lepidium didymum*, *Amaranthus powellii*, and *Lolium multiflorum* seedlings.

Trial	Species	Mean Size	Treatments
1	<i>L. didymum</i>	Stem length 64.0 mm (SD = 13.9 mm) Stem basal diameter 1.9 mm (SD = 0.5 mm)	Plants grown: in bags vs in ground Application: to leaves pressed to dry soil surface Dose applied: no treatment vs 25, 50 or 100 x 100 $\mu$ s pulse lengths at 4.5 kV with electrode disc pressing plant to soil. Extra treatments: inserted rod vs surface pressed disc earthing electrode applying 100 x 100 $\mu$ s pulses at 4.5 kV with different electrode separation distances.
2	<i>A. powellii</i>	Stem length 72.9 mm (SD = 12.3 mm) Stem basal diameter 2.1 mm (SD = 0.3 mm)	Plants grown: in bags vs in ground Application: to leaf canopy only vs leaves pressed to dry soil surface Dose applied: no treatment vs 25 x 25 $\mu$ s, 50 x 50 $\mu$ s, 50 x 100 $\mu$ s, 100 x 100 $\mu$ s, or 100 x 200 $\mu$ s pulses at 4.5 kV.
3	<i>L. multiflorum</i>	Tiller No. 1.2 (SD = 0.3) Leaf No. 2.9 (SD = 0.5) Longest leaf length 157.6 mm (SD = 17.1 mm)	Plants grown: in bags vs in ground Application: to leaf canopy only vs leaves pressed to dry soil surface Dose applied: no treatment vs 100 x 200 $\mu$ s pulses, 200 x 200 $\mu$ s pulses and 200 x 400 $\mu$ s pulses at 3.5 kV or 4.5 kV.

For *L. multiflorum*, data were analysed with each bag of three grasses representing one replicate (n = 6). Following the recommendations of Armstrong (2014) and Perneger (1998), no multiple test (e.g., Bonferroni) corrections were applied. The Kruskal-Wallis pairwise and multiple comparisons stepwise step-down procedures were used to determine homogeneous sets. For both *A. powellii* and *L. multiflorum*, binary logistic regression was used to assess the factors contributing to plant death. The *L. multiflorum* binomial logistic regression analysis used data for individual plants rather than mean values of bags of three plants and excluded controls.

## 5.5 Results

### 5.5.1 *Lepidium didymum*

When bag grown *L. didymum* plants were treated with the disc earthing electrode, there was evidence of arcing with the treatment electrode disc, indicated by loud cracking and sparking observed during treatment, so the energy applied to some plants may have been very little or none. Assessment of plant vigour 7 DAT showed all untreated control plants and some disc-earthed plants were healthy with a plant vigour score = 1.0 (Figure 5-4).

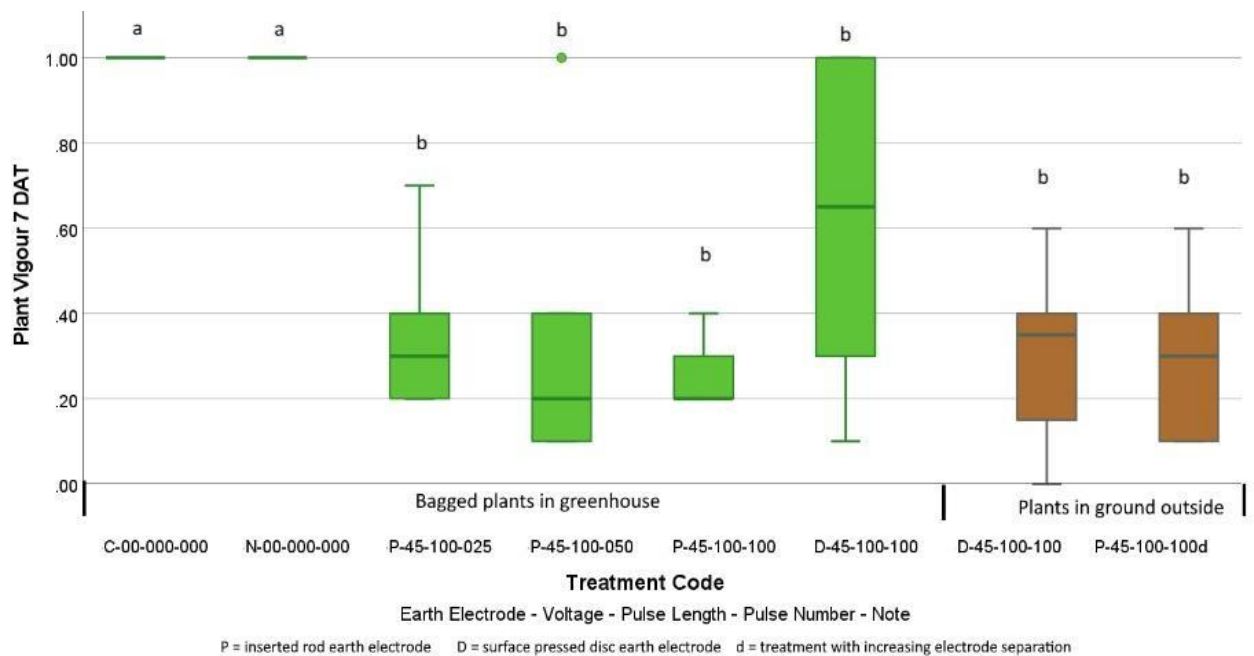


Figure 5-4 Clustered boxplot of plant vigour score (0 = dead, 1 = healthy) 7 days after treatment by treatment code for *Lepidium didymum* in bags indoors and outdoors in the ground. A mild outlier is shown by a green dot. Plots sharing the same letters are not significantly different as determined using Kruskal-Wallis pairwise multiple comparisons.

Indeed, most of the bag-grown, disc-earthing electrode treatment plants were healthy, giving a mean plant vigour score = 0.62. The outdoor grown plants treated using the disc earthing electrode were much weaker with a vigour score = 0.28. Vigour of the probe earthing electrode treatment plants were 0.28 for both outdoor grown treatments, 0.25 for

the highest indoor treatment and 0.35 and 0.33 for the lowest and middle treatment, respectively. Kruskal-Wallis pairwise multiple comparisons showed all treatments had significantly reduced vigour compared to the controls, but there was no significant difference among the treatments (Figure 5-4). After 27 days, none of the untreated control plants had died, and all in-ground treated plants had died. All the greenhouse bag-grown plants treated with the inserted-probe earthing electrode died but half the disc earthing electrode treated plants, where probable arcing was noted, survived. For the indoor-grown bagged plants, the amount of energy discharged using the inserted rod earthing electrode increased exponentially as the duration of discharge doubled (Table 5-2).

Table 5-2 Mean energy discharge for Trial 1 when *Lepidium didymum* growing in bags or in the ground were treated with 4.5 kV x 100 µs pulse doses using either a probe or a disc earthing electrode. Mean energy discharge values sharing the same letters are not significantly different as determined using Kruskal-Wallis pairwise multiple comparisons.

<b>Planting</b>	<b>Earthing</b>	<b>Voltage</b>	<b>Pulse Length (µs)</b>	<b>Number of Pulses</b>	<b>Mean Energy Discharge (J)</b>
<b>Bagged</b>	Probe	4,500	100	25	4.6 <sup>b</sup>
<b>Bagged</b>	Probe	4,500	100	50	8.4 <sup>c</sup>
<b>Bagged</b>	Probe	4,500	100	100	23.0 <sup>d</sup>
<b>Bagged</b>	Disc	4,500	100	100	4.5 <sup>b</sup>
<b>In-ground</b>	Disc	4,500	100	100	2.2 <sup>a</sup>

Comparing 4.5 kV, 100 x 100 µs pulses, the energy discharge using a disc electrode pressing plants against a dry soil surface was approximately 20% that of the same dose applied with an inserted rod earthing electrode. The energy discharge of applications with a disc earthing electrode pressed to soil in the open ground was 48% of that when the

same dose was applied in bags. The energy discharged reduced with increasing electrode separation distance, exhibiting a strong decay curve (Figure 5).

Power function trendlines showed a strong correlation for the applications made with a plant pressed to the soil surface ( $R^2 = 0.99$ ) and the applications to a bare soil surface only ( $R^2 = 0.92$ ). The power function was greater for the applications including plants, and discharges were 2.5 times greater for applications made to plants and soil than those made to the soil only.

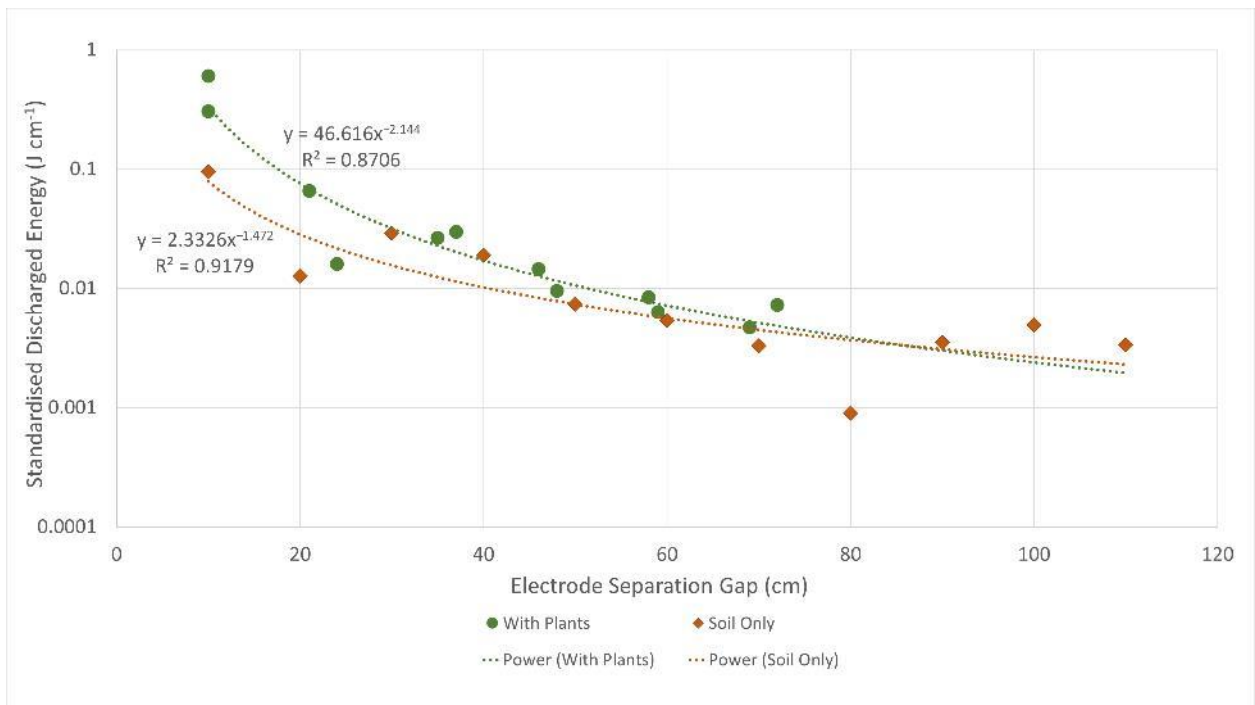


Figure 5-5 Logarithmic scatterplot of standardized discharged energy ( $J\ cm^{-1}$ ) showing the correlation with power functions of discharged energy by electrode separation gap with the treatment disc-electrode pressing *Lepidium didymum* plants to the soil surface (green circles) or pressed to bare soil only (brown diamonds).

### 5.5.2 *Amaranthus powellii*

All untreated *A. powellii* control plants were alive and healthy 12 DAT but there was a general trend for a lower condition score and a higher death rate with higher energy treatments (Figure 5-6). There was little difference between the bag grown plants treated

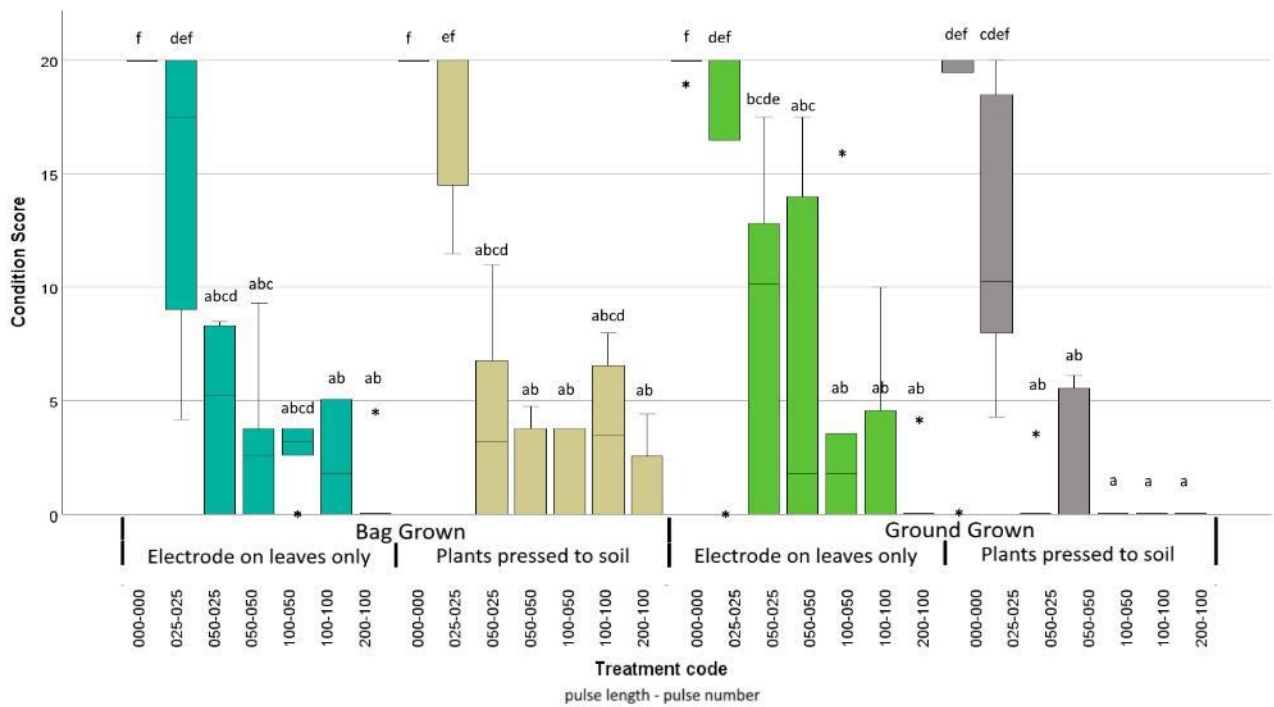


Figure 5-6 Clustered boxplot of *Amaranthus powellii* plant condition 12 days after treatment with pulsed microshocks by treatment showing that untreated plants were healthy, and that the condition of treated plants reduced with increasing electrical dose applied. Plots sharing the same letters are not significantly different as determined using Kruskal-Wallis pairwise multiple comparisons.

by the electrode applied only to the leaf canopy or pressing the plant to soil. In-ground plants with treatments applied to the leaf canopy were generally healthier, whereas there were more early deaths among plants treated by pressing the whole plant to the soil.

At 15 DAT, all the *A. powellii* untreated control plants were healthy, and in all treatments, there were some survivors and some deaths at the lowest energy doses. At higher doses, all plants died (Figure 5-7). Far more energy was discharged when equivalent treatments were applied to *A. powellii* plants growing in bags than to plants growing in the ground

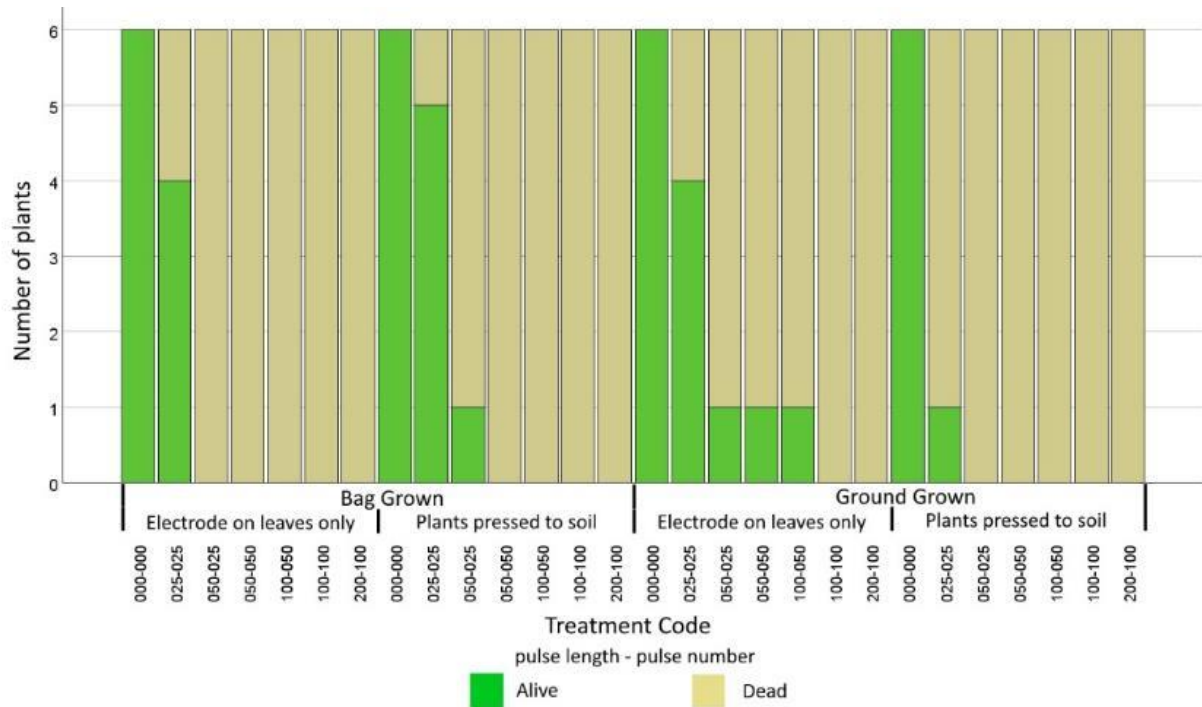


Figure 5-7 Histogram of *Amaranthus powellii* plants surviving 15 days after treatment comparing bagged versus in-ground plants, and the flat plate electrode contacting the leaves only versus the electrode pressing plants to the soil. All untreated plants (000-000) survived, some plants given lower energy treatments survived, but all plants receiving the higher doses were killed.

and more energy was discharged when applying treatments to whole plants pressed to the soil surface than to leaves only. Comparing equivalent doses applied under different scenarios showed that leaf-only treated plants in the ground used an average of 14.8% of the energy of leaf-only treated plants in bags, and soil-pressed treated plants in the ground used 24.2% of the energy of soil-pressed treated plants in bags. Leaf-only treated plants in bags used 62.3% of the energy of soil-pressed treated plants in bags, and leaf-only treated plants in the ground used 49.7% of the energy of soil-pressed treated plants in the ground (Figure 5-8).

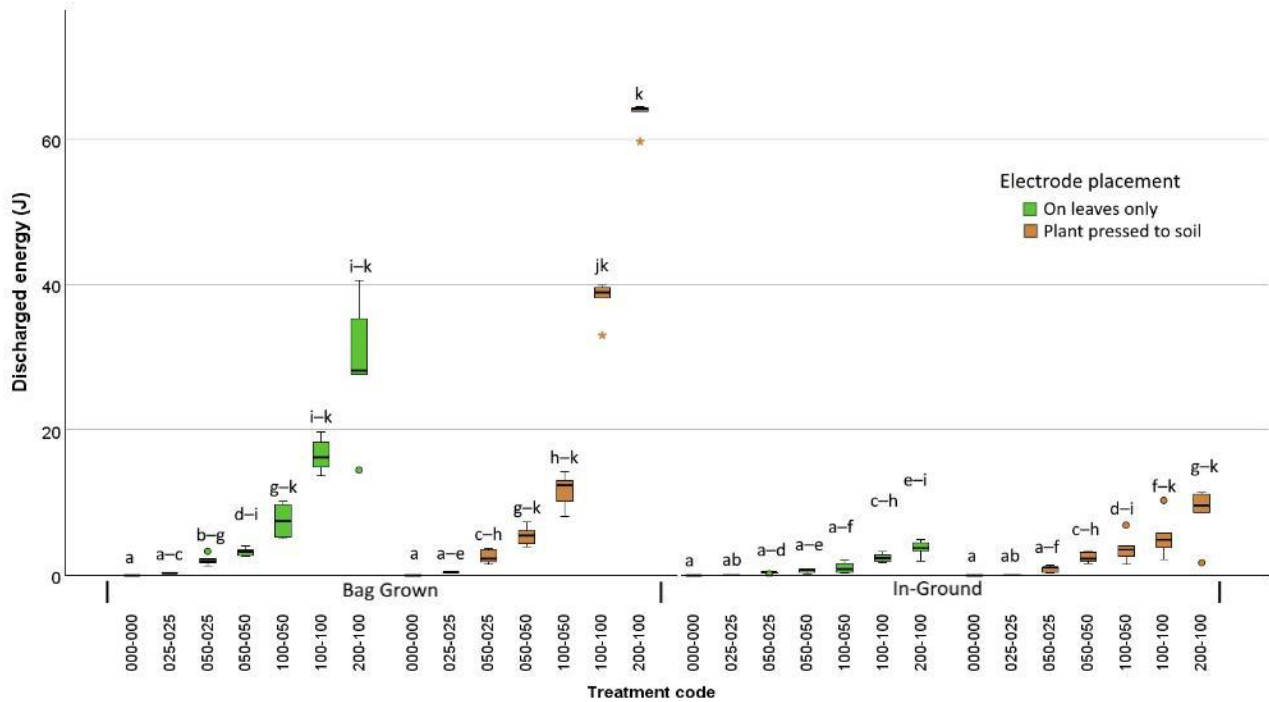


Figure 5-8 Clustered boxplot of discharged energy by treatment applied to *Amaranthus powellii* in bags and in the ground with the treatment electrode either pressing against the leaves only or pressing the plant to the soil surface. Mild outliers are indicated by dots and extreme outliers are indicated by stars. Plots sharing the same letters are not significantly different as determined using Kruskal-Wallis pairwise comparisons.

To compare the discharge rate of different treatments, data were standardised by dividing measured discharge by the actual on-time (pulse length x pulse number) to determine the discharge rate as kilojoules per second ( $\text{kJ s}^{-1}$ ) which is equivalent to power in kilowatts (kW). The discharge rate of the shortest duration treatment (25 x 25  $\mu\text{s}$  pulses) was much lower than the average rate for longer duration treatments (Table 5-3). Data are presented in Table 5-4 to show the mean discharge rate ( $\text{kJ s}^{-1}$ ) for all dose durations (labelled  $> 0$  ms), the mean discharge rate for the shortest dose duration (0.625 ms) and the mean discharge rate for the average of all longer dose durations (labelled  $> 0.625$  ms). With one bag-grown exception, all plants for which the energy discharge exceeded 0.5 J died (n=108) and more than 90% of plants treated with more than 0.3 J died.

Table 5-3 Table showing the mean energy discharge rate ( $\text{kJ s}^{-1}$ ) by dose applied (voltage-pulse length-pulse number) for *Amaranthus powellii* plants growing in bags or in the ground and the treatment electrode pressed to the leaves only or pressing the plant against the soil surface.

Mean energy discharge ( $\text{kJ s}^{-1}$ )				
Treatment	Bag-grown		Ground-grown	
Dose Applied	Leaves only	Pressed to Soil	Leaves only	Pressed to Soil
45-025-025	0.518	0.667	0.053	0.050
45-050-025	1.674	2.022	0.303	0.724
45-050-050	1.302	2.178	0.252	0.976
45-100-050	1.505	2.345	0.207	0.740
45-100-100	1.649	3.807	0.241	0.529
45-100-100	1.452	3.170	0.1851	0.459

Table 5-4 Comparisons of *Amaranthus powellii* seedling plants treated in bags and in-ground, with the treatment electrode applied to the leaves only or pressing the plant against the soil surface, showing the mean energy discharge per plant (J), mean energy discharge rate ( $\text{kJ s}^{-1}$ ) and mean plant death rate 15 days after treatment

	Dose discharge duration (ms)	Bag Grown Plants			In-ground Grown Plants		
		> 0	0.625	>0.625	> 0	0.625	>0.625
<b>Average of all plants</b>	Mean death rate (%)	86.1	25.0	98.3	88.7	58.3	94.9
	Mean energy discharge/plant (J)	12.9	0.37	17.9	2.01	0.032	2.82
	Mean energy discharge rate ( $\text{kJ s}^{-1}$ )	1.86	0.59	2.11	0.391	0.052	0.459
<b>Electrode contacting leaf canopy only</b>	Mean death rate (%)	88.9	33.3	100	80.6	33.3	90.0
	Mean energy discharge/plant (J)	8.39	0.32	11.7	1.17	0.033	1.63
	Mean energy discharge rate ( $\text{kJ s}^{-1}$ )	1.35	0.51	1.52	0.207	0.053	0.238
<b>Electrode pressing whole plant to soil</b>	Mean death rate (%)	83.3	16.7	96.7	97.1	83.3	100
	Mean energy discharge/plant (J)	17.4	0.41	24.2	2.87	0.031	4.21
	Mean energy discharge rate ( $\text{kJ s}^{-1}$ )	2.36	0.66	2.70	0.576	0.050	0.681

An example of pulse data captured by the WedaTech equipment during a single PMS application to *A. powellii* is shown in Figure 5-9. This shows an initial drop in voltage and rise in current as the treatment begins, followed by a relatively stable voltage with the current continuing to increase during the treatment. This pattern was typical of a higher energy treatment when good electrode contact with the plant was maintained.

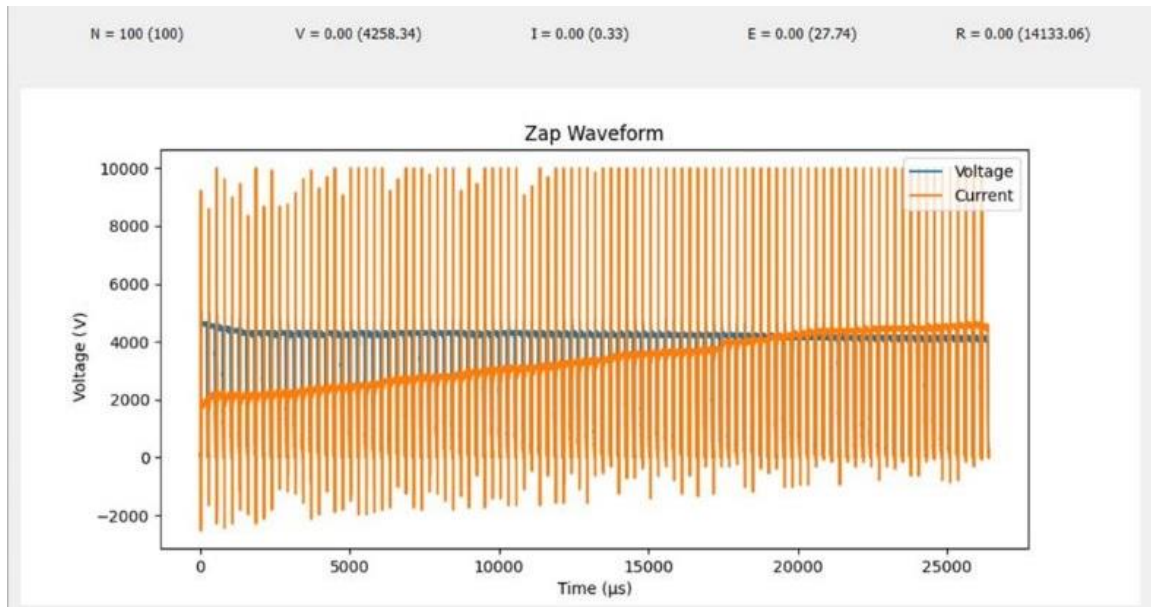


Figure 5-9 Screenshot of the WedaTech Zapper control interface showing the set number of pulses, voltage, time per pulse, period of pulses and a chart of the resulting oscilloscope measurements of voltage and current during application of a treatment to the upper leaves of an *Amaranthus powellii* seedling. N = logged number of pulses, V = mean voltage, I = mean current (amps), E = calculated energy (Joules), and R = calculated mean resistance (Ohms).

I also noted that if good contact with the plant was not achieved, the current could reduce markedly. Figure 5-10 shows a case where initial arcing was indicated by sparking/crackling noises, and the flat plate electrode was adjusted to better contact the leaves. While the voltage remained constant, the current initially dropped, but then climbed once good contact was achieved, following a similar trend to that observed with other applications made using similar doses.

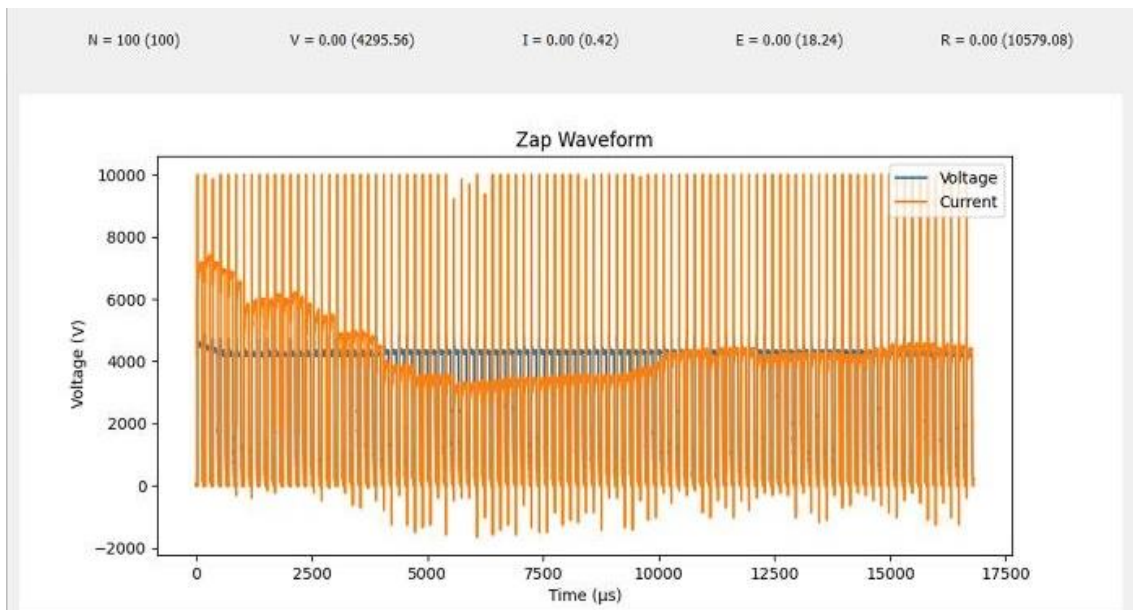


Figure 5-10 Screenshot of the WedaTech Zapper control interface showing a chart of the resulting oscilloscope measurements of voltage and current during application of a treatment to the upper leaves of an *Amaranthus powellii* seedling with poor initial contact.

Logistic regressions were performed to ascertain the effects of treatment on the likelihood that plants would be killed (Table 5-5). Included variables were electrode contact to leaves only or to leaves pressed to soil, soil moisture level, stem length and stem diameter at time of treatment, and average treatment voltage, current and discharged energy. The logistic regression model was statistically significant,  $\chi^2(7) = 124.1$ ,  $p < 0.001$ . The model explained 77.3% (Nagelkerke  $R^2$ ) of the variance in the outcome and correctly classified 91.1% of the cases. Increasing average voltage ( $\text{Exp}(B) = 1.001$ ,  $p = 0.005$ ), average current ( $\text{Exp}(B) = 0.000$ ,  $p = 0.026$ ), and discharged energy ( $\text{Exp}(B) = 21.08$ ,  $p = 0.005$ ) were significant factors. Soil moisture ( $\text{Exp}(B) = 1.296$ ,  $p = 0.069$ ) and the other variables were not significant.

Table 5-5 Variables entered in the equation for a binary logistic regression for predicting the likelihood that *Amaranthus powellii* seedlings treated with pulsed electric shocks would be killed.

Variables in the Equation		95% C.I. for EXP(B)							
		B	S.E.	Wald	df	Sig.	Exp(B)	Lower	Upper
Step 1	Electrode Contact	1.116	.784	2.028	1	.154	3.054	.657	14.193
	Soil Moisture (%)	.259	.143	3.303	1	.069	1.296	.980	1.715
	Stem Length (mm)	-.030	.034	.799	1	.371	.970	.908	1.037
	Stem Diameter (mm)	-.133	1.258	.011	1	.916	.875	.074	10.300
	Mean Voltage (V)	.001	.000	7.965	1	.005	1.001	1.000	1.001
	Mean Current (I)	-8.397	3.765	4.974	1	.026	.000	.000	.362
	Discharged energy (J)	3.048	1.076	8.023	1	.005	21.080	2.557	173.768
	Constant	-8.436	4.435	3.618	1	.057	.000		

### 5.5.3 *Lolium multiflorum*

None of the untreated *L. multiflorum* control plants had died 14 DAT, but some plants had died in all the treatments and more than 90% of plants were dead in 16 of the 28 treatments. Some treated plants that were initially identified as dead resprouted from underground buds. The bag-grown plants had a higher mean survival rate than the in-ground plants, but this was not statistically different when analysed using Kruskal-Wallis all-pairwise comparisons (Figure 5-11).

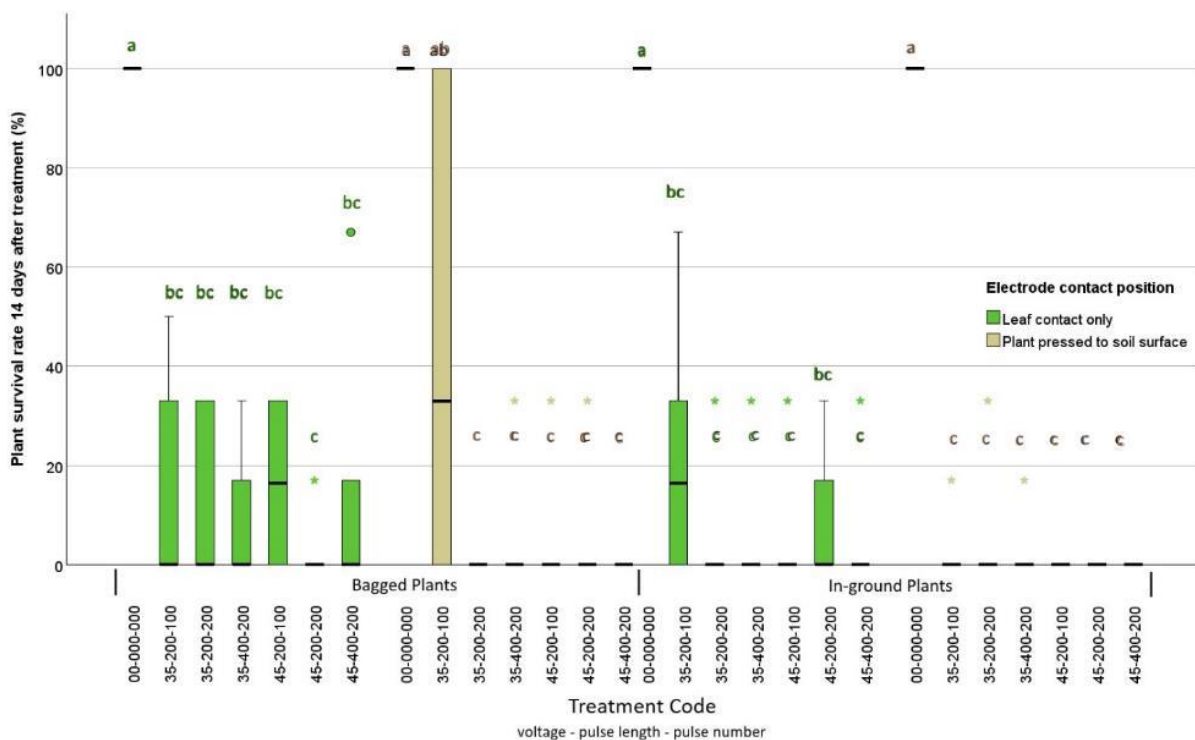


Figure 5-11 Clustered boxplot of *Lolium multiflorum* plant survival rate 14 days after treatment by dose applied to plants in bags and in the ground with the treatment electrode either pressing against the leaves only or pressing the plant to the soil surface. A mild outlier is indicated by a green dot and extreme outliers by stars. Plots sharing the same letters are not significantly different using Kruskal-Wallis all-pairwise comparisons.

Comparing the survival rate of plants with the energy discharged showed that death rate tended to increase with increasing energy (Figure 5-12). The discharged energy for each application was analysed and results for bagged or in-ground plants, electrode contact position and applied dose were compared (Figure 5-13).

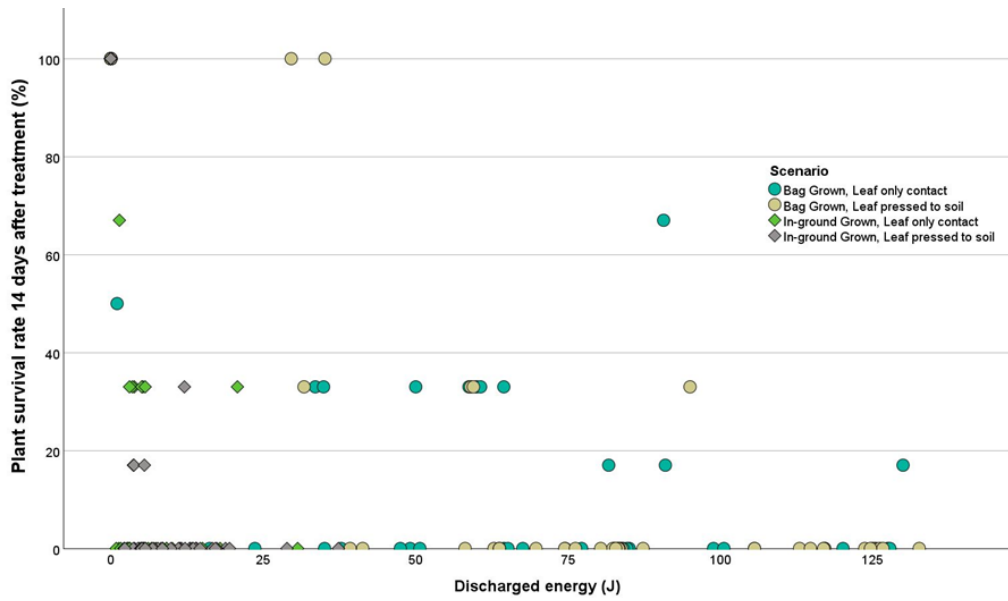


Figure 5-12 Scatterplot of *Lolium multiflorum* mean survival rate per bag 14 DAT by discharged energy for bag-grown and in-ground plants and both leaf-only or leaf pressed to the soil surface treatments. Each discharge treated three *L. multiflorum* seedlings at the same time.

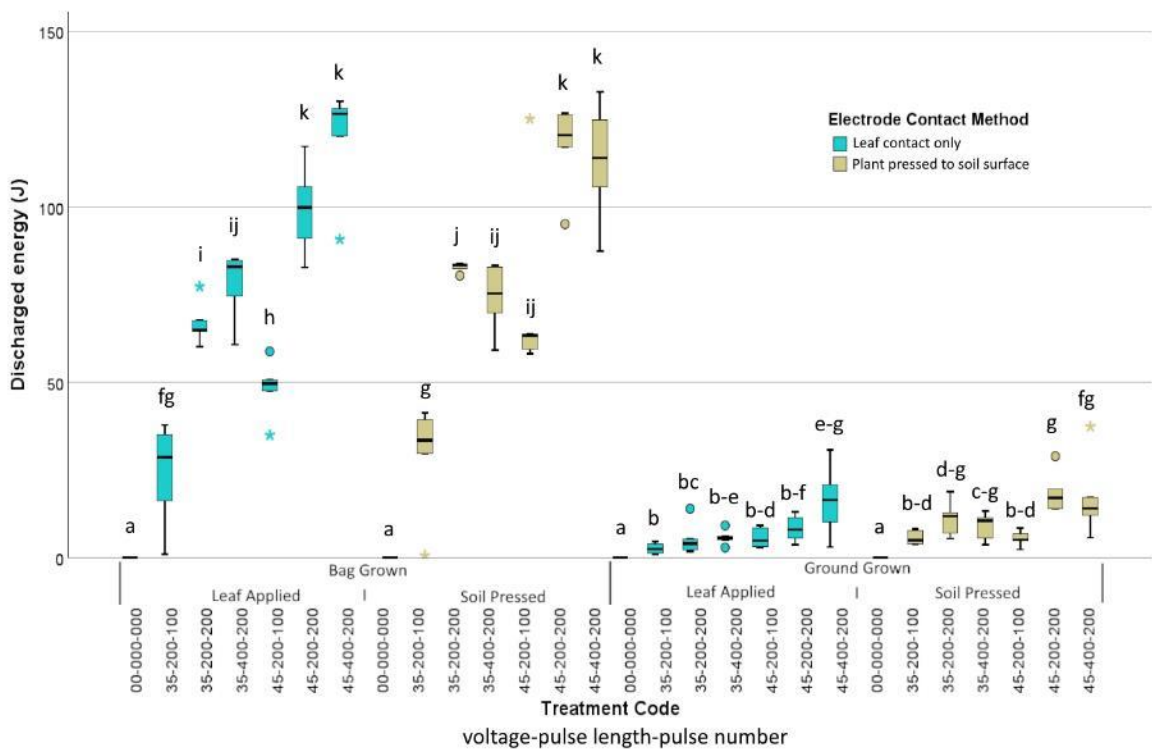


Figure 5-13 Clustered boxplot of discharged energy for treatments applied to *Lolium multiflorum* in bags and in the ground. Mild outliers are indicated by dots and extreme outliers by stars. Plots with the same letters are not significantly different as determined using Kruskal-Wallis Stepwise-Step Down multiple comparisons.

An extreme outlier in the B-S-35-200-100 treatment (bagged plants pressed to the soil applying 3,5 kV with 100 pulses each of 200  $\mu$ s) was identified in post-treatment electrical data files as a failed application with very low energy applied, and mean energy for the treatment was substituted. A second relatively low energy application was retained, and those plants had a higher survival rate than others in the treatment. There were also low-energy outliers in other treatments, but they were retained as a relatively high amount of energy was discharged, even if not at the same rate as others in the same treatment. An extreme higher-energy discharge in treatment B-S-45 200-100 was noted as having arced to the ground, but data were retained in further analyses as at least the target dose was applied.

On an individual plant basis, 87.5% of bag grown plants died at a mean energy discharge of 77.3 J. This equates to 3.86 MJ ha<sup>-1</sup> assuming 5 escape weeds ha<sup>-1</sup> (Table 5-6).

Table 5-6 Summary of the death rate and discharged energy when *Lolium multiflorum* plants in bags or in the ground were treated with the treatment electrode applied to the leaf canopy only or to whole plant pressed to the soil surface at either 3.5 or 4.5 kV.

	Voltage (kV)	Bag Grown Plants			In-ground Plants		
		3.5	4.5	All	3.5	4.5	All
	Death Rate (%)	83.3	91.7	87.5	91.7	96.3	94.0
<b>All plants</b>	Energy Discharge (J)	59.4	95.2	77.3	6.58	11.8	9.18
	Energy ha <sup>-1</sup> (MJ ha <sup>-1</sup> ) *	2.97	4.76	3.86	0.329	0.589	0.507
	Death Rate (%)	87.0	87.0	87.0	88.9	92.6	90.7
<b>Leaf canopy only contacted</b>	Energy Discharge (J)	56.5	89.4	73.0	4.49	10.0	7.26
	Energy ha <sup>-1</sup> (MJ ha <sup>-1</sup> ) *	2.82	4.47	3.65	0.224	0.502	0.363
	Death Rate (%)	79.6	96.3	88.0	94.4	100	92.7
<b>Leaves pressed to soil</b>	Energy Discharge (J)	62.2	101	81.6	8.68	13.5	11.1
	Energy ha <sup>-1</sup> (MJ ha <sup>-1</sup> ) *	3.11	5.05	4.10	0.434	0.675	0.555

The death rate and energy discharge were higher at 4.5 kV than at 3.5 kV. When only the in-ground plants are considered, the plant death rate was higher at 94% and the energy to kill plants was much lower with a mean energy discharge of 9.18 J application<sup>-1</sup> or 0.051 MJ ha<sup>-1</sup>. The death rate and energy discharge were higher for 4.5 kV treatments than for 3.5 kV treatments, and for plants when the electrode pressed the leaves to the soil surface rather than to the leaves only.

A logistic regression was performed to ascertain the effects of treatment on the likelihood that plants would be killed (Table 5-7). Included variables were electrode contact to leaves only or to leaves pressed to soil, soil moisture level, leaf number and longest leaf length at time of treatment, and average treatment voltage, current and discharged energy. The logistic regression model was statistically significant,  $\chi^2(7) = 160.9$ ,  $p < 0.001$ . The model explained 48.0% (Nagelkerke  $R^2$ ) of the variance in the outcome and correctly classified 91.9% of the cases. However, the Hosmer and Lemeshow test showed very poor model fit ( $p < 0.001$ ). Applying the electrode to leaves pressed to the soil surface, (Exp(B) = 2.208,  $p = 0.023$ ), fewer leaves (Exp(B) = 0.603,  $p = 0.038$ ) and longer leaves (Exp(B) = 1.001,  $p = 0.034$ ) at time of treatment, average voltage (Exp(B) = 1.001,  $p < 0.001$ ) and discharged energy (Exp(B) = 1.027,  $p = 0.002$ ) significantly increased likelihood of death. Higher percentage soil moisture (Exp(B) = 0.975,  $p = 0.361$ ) and average current (Exp(B) = 0.064,  $p = 0.501$ ) were not significant.

Table 5-7 Variables entered in the equation for a binary logistic regression for predicting the likelihood that *Lolium multiflorum* seedlings treated with pulsed electric shocks would be killed.

Variables in the Equation	95% C.I. for EXP(B)							
	B	S.E.	Wald	df	Sig.	Exp(B)	Lower	Upper
Electrode Contact	.792	.349	5.150	1	.023	2.208	1.114	4.375
Leaf number	-.506	.244	4.290	1	.038	.603	.374	.973
Longest leaf (mm)	.014	.007	4.509	1	.034	1.014	1.001	1.028
Soil moisture (%)	-.025	.028	.834	1	.361	.975	.924	1.029
Step 1 Mean Voltage (V)	.001	.000	58.849	1	<0.001	1.001	1.001	1.001
Mean Current (I)	-.690	1.04 9	.433	1	.510	.501	.064	3.916
Discharged energy (J)	.026	.009	9.370	1	.002	1.027	1.010	1.044
Constant	-2.334	1.31 1	3.172	1	.075	.097		

## 5.6 Discussion

Seeking a non-chemical herbicide method suitable for use in a regenerative agriculture cropping system, these trials compared the effects of low-energy electric shocks on two broadleaf weed species and a grass, treated both in bags and grown in the ground in a cropping field.

In the *L. didymum* trial, all treated plants and no untreated control plants died, although due to slow death rates it was almost 4 weeks from treatment before final determinations could be made. A discharge on 0.3 J or less appears sufficient to kill small *L. didymum* seedlings. Plants grown in bags and held indoors died faster than those growing outside in the ground. Even the lowest energy treatment (2.5 ms total discharge duration) applied

to bagged plants had a greater energy discharge than any treatment applied to plants or bare soil in the ground (10 ms total discharge duration).

The doses applied to *A. powellii* were selected to span treatments expected to be ineffective through to those expected to be fully effective, but in this trial even the lower doses were very damaging to the weeds. Any *A. powellii* seedlings receiving a dose of 0.5 J plant<sup>-1</sup> were killed, and more than 90% of plants were killed with a dose as low as 0.3 J plant<sup>-1</sup>. While two of my earlier reported trials (Bloomer et al., 2022, 2023a) had only moderate success killing *L. multiflorum* plants grown in bags, this trial demonstrated a very high death rate, with over 94% of in-ground treated plants and 87.5% of in-bag plants dying. The plants in this trial were slightly larger than in two earlier trials (Chapter 3 (Bloomer et al., 2022)) but slightly smaller than those in the two other reported trials (Chapter 4 (Bloomer et al., 2023a)) (Table 5-8). On an individual plant basis, when only the in-ground grown plants are considered, plant death rate was 94% and the energy to kill plants is much lower with a mean energy discharge of 3.06 J plant<sup>-1</sup>.

Table 5-8 Comparison of key plant measurements for *Lolium multiflorum* in successive experiments showing that seedlings in the current trial were of similar size to earlier trials.

<b>Trial</b>	<b>Tiller No.</b>	<b>Leaf No.</b>	<b>Longest leaf length</b>
<b>Current</b>	1.2	2.9	158 mm
<b>Previous [A] 1*</b>	1.0	2.0	109.mm
<b>Previous [A] 2^</b>	1.0	2.0	149 mm
<b>Previous [B] 1^</b>	1.6	3.7	141 mm
<b>Previous [B] 2^</b>	1.9	4.0	197 mm

[A] (Bloomer et al., 2022)      [B] (Bloomer et al., 2023a)  
 \* single pulse treatments      ^ multiple pulse treatments

For *A. powellii* seedlings, almost seven times more energy was discharged to bag-grown plants than to in-ground plants when treatments contacted the leaf canopy only. Four times more energy was discharged to bag-grown plants than to in-ground plants when treatments pressed the plants to the soil surface. In both the bag and ground grown plants, treatments that pressed plants to the soil surface experienced about twice the energy discharge of equivalent leaf canopy only doses. How much of the difference was due to better electrode-to-leaf contact and how much was due to direct energy loss from the flat plate electrode to the soil could not be determined. In this trial, the 4,500 V treatments on *L. multiflorum* were more effective than 3,500 V treatments, although energy discharge was also much higher. Overall, pressing the grasses to the ground had a slightly higher kill rate than touching the plate electrode to the leaves only. When treating *L. multiflorum* seedlings, the energy discharged to bag-grown plants with significantly wetter soil was much greater than that discharged to in-ground plants at the same machine settings, with bagged plant treatments discharging ten times more energy for leaf only treatments and seven times more for soil pressed treatments, but this did not necessarily correlate with a higher death rate.

Ohm's Law for the calculation of circuit resistance (R) from the measured voltage (V) and current (I) (arranged as Equation 5-1) states that resistance in a circuit is a function of voltage and current.

$$R = V / I \qquad \text{Equation 5-1}$$

The initial drop in voltage and rise in current as treatment of *A. powellii*, followed by a relatively stable voltage but the current continuing to increase during treatment (Table 5-3 and Figure 5-9) show that resistance was decreasing during application, initially quite quickly and then more slowly as the pulses continued to be applied.

This supports hypotheses that the electric shocks are increasing the conductivity of plant tissues, perhaps by damaging the cell membranes and releasing more ionic fluids into the intercellular spaces (Mizuno et al., 1990; Gusbeth et al., 2004; Baev & Yudaev, 2017; Yudaev, 2019). Therefore, the lower energy discharge rate ( $\text{J sec}^{-1}$ ) or power (watts, W) observed in the shorter duration treatments applied to *A. powellii* is indicative of higher initial vegetative resistance. Conversely, during the longer duration applications more of the pulses would be treating damaged tissues with reduced resistance. This raises the possibility that monitoring resistance during PMS application may enable treatment to be stopped when sufficient damage has occurred to ensure subsequent plant death. Figure 5-10 presents an example in which current initially decreased, showing that initial resistance increased. When the treatment electrode contact with the canopy was improved part way through the application, the current increased showing the resistance decreased because voltage remained constant. This demonstrates that good electrode connection with the plant is essential for the potential dose of energy to be applied.

The design of my application equipment means that, up to a system limited maximum discharge current, the energy discharge ( $E_p$ ) is controlled by the voltage (V), the total discharge time ( $t_c$ ) and the resistance of the plant/soil machine circuit ( $R_v$ ). Slaven et al. (2023) relate these factors in the formula presented as Equation 5-2.

$$E_p = \frac{V^2 \times t_c}{R_v} \quad \text{Equation 5-2}$$

Other factors that can influence energy discharge include electrical properties of epidermal tissues, the cuticle and waxy layers, the initial hydration status of the plant and physical structure variation. Equation 5-2 does not explicitly identify soil resistance, which is a critical factor because it is a significant part of the electrical circuit for plants

treated when growing in the ground. While good earthing electrode connection and good treatment electrode connection to the plant are essential for effective treatment, direct connection between the treatment electrode and the soil body creates inefficiency. When the flat plate presses the plants to the soil surface, there will be electrical conductance into the soil both through the plant and from direct electrode to soil contact. The energy split will depend on the relative resistance of the two paths, and anything that increases resistance between the plate electrode and the soil will increase the energy available for discharge through the plant. The data presented in Figure 5-5 show that flat plate discharges were 2.5 times greater for applications made to plants and soil than those made to the soil only, indicating much higher conductivity of plants relative to the soil surface.

The resistance of the soil is dependent on several factors, including soil physical properties such as clay content and density, variable factors including salinity and in my trials, electrode surface contact area, electrode separation distance, and soil moisture, which all affect soil electrical conductivity (McNeill, 1980). A larger electrode surface area or higher soil moisture will reduce resistance whereas increasing electrode separation distance increases resistance. Diprose and Benson (1984) state that high soil resistance can leave less capacity to shock the plants. Discussing broadacre application equipment, they recommended that earthing discs penetrate several centimetres into the ground and have large cross-sectional area to ensure adequate electrical contact. Pre-trial testing of my equipment showed that pairing fully embedded 200 mm long, 5 mm diameter, aluminium electrodes in wet soil allowed the current between them to exceed the measuring capacity of my equipment, indicating that electrical resistance was extremely low. I believe the very low resistance was due to both the larger contact area and the much

wetter soil below about 100 mm. Inserting the electrodes only 75 mm into the soil reduced the current and enabled my equipment to capture reliable measurements.

During the *L. didymum* trial in the field, I tested the effect of increasing the electrode separation gap, and of two earthing electrode types on energy discharge. Hydrosense II™ soil moisture measurements around the outdoor grown plants identified a moisture gradient down the soil profile, with the surface layer air dry and the deeper layers very moist. Under this scenario, there was an apparent effect of electrode separation distance on discharge rate, which can be assumed to be a function of soil resistance. A power-function decay was observed (Figure 5-5), showing a rapid decrease in energy discharge up to about 30 cm electrode separation beyond which the discharge decrease was minimal.

The average soil moisture content for the *L. didymum* trial was similar for bagged and in-ground plants, but the in-ground plants had a dry surface soil layer, and the in-ground conditions were more variable than within the bags, particularly with increasing soil depth. At equivalent machine settings, the higher energy discharge with the probe earthing electrode compared to the disc earthing electrode, and the higher energy discharge with plants present than without, suggested the dry soil surface acts as an electrical insulator.

Diprose et al. (1978) suggested that much more energy would be required to achieve satisfactory kill rates of in-ground plants than bag-grown plants in a greenhouse and noted that “effects are variable depending on plant root type, age, size, and the relative moisture contents of the soil”. While more energy was discharged to bag-grown plants, I did not find that more energy was required to successfully control weeds growing in the ground

relative to that required to kill bag-grown plants, all other things remaining equal. My trials do indicate that plant size and development are important, especially for *L. multiflorum* seedlings once tillering has commenced, and the method of electrode earthing and treatment electrode placement on plants also impacts treatment effectiveness. Binomial logistic regression of data from the *A. powellii* and *L. multiflorum* trials sought to identify the role various factors had on the likelihood that seedlings would be killed. While soil moisture can have a large impact on the amount of energy discharged, it was not necessarily related to the probability that *A. powellii* or *L. multiflorum* seedlings will be killed. Increasing voltage and increasing the energy discharged were significant predictors of the likelihood of treatments killing seedlings of both species.

I set a goal of attaining the energy efficiencies achieved in my earlier greenhouse trials, with better than 90% control using energy of less than 1 MJ ha<sup>-1</sup> plus transport at 5 weeds m<sup>-2</sup>, which is considered a reasonable density for escape weeds surviving after chemical or mechanical treatment. Better than 90% control was achieved for both *L. didymum* and *A. powellii* at 0.3 J plant<sup>-1</sup> which equates to 15 kJ ha<sup>-1</sup>, and for in-ground *L. multiflorum* at 363 kJ ha<sup>-1</sup> (leaf contact) and 555 kJ ha<sup>-1</sup> (leaves pressed to soil), all well below my target of 1 MJ ha<sup>-1</sup>. These energy efficiencies are equivalent to or better than my previously published greenhouse trials as reported in Chapters 3 and 4 (Bloomer et al., 2022, 2023a).

The equipment we developed and tested is suited to treating individual weeds or small clumps of weeds up to about 15 cm tall. The use of a flat plate electrode is proven to be effective and efficient in a field setting. The system would fit well with robotic “spot

weeders” equipped with weed recognition which is increasingly available (Ziwen et al., 2015; Ahmad et al., 2018; Li et al., 2019; Anken & Latsch, 2022; Coleman, Bender, et al., 2022; Coleman & Salter, 2023). The flat plate electrode requires accurate placement which could be achieved by robots such as BoniRob (Michaels et al., 2015), or other systems using using the delta arm configuration (Malewar, 2021; Nguyen & Tung, 2024) or other accurate systems (Lysakov et al., 2021). In a small unreported trial I applied the circular disc treatment electrode to newly germinated seedlings growing very closely together. With thin insulation on the side of the disc, the plants immediately adjacent to those being treated were unaffected (Figure 5-14).

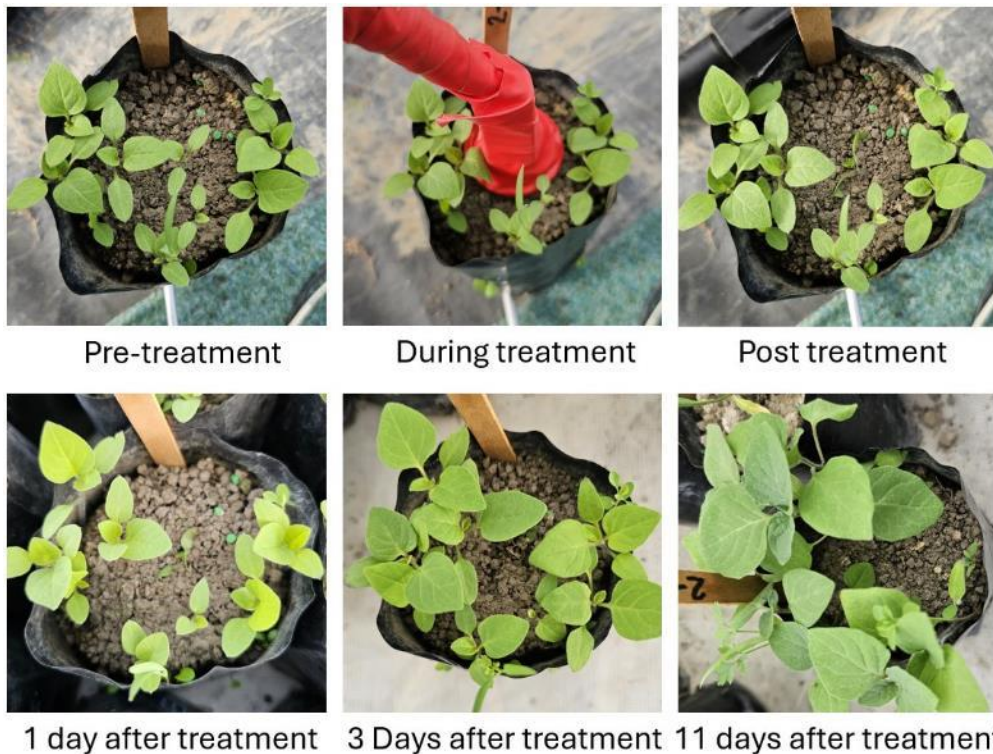


Figure 5-14 Time series photographs taken before during and immediately after treatment with pulsed electric microshocks of seedlings surrounding a central weed, and at 1, 3 and 11 days after treatment was applied showing adjacent weeds were unaffected.

Being an ultra-low energy method, PMS would be ideal for low powered robots including those using only solar power such as the early Ecorobotix machine (Vaqr, 2018;

Abdelghafour et al., 2021). The electric weeding system does require adequate earthing, which may reduce ease of use in a crop with a high amount of residue cover. A flat plate system can press onto such residues, and if a metal disc cutting into the ground is blocked by debris, a second probe spearing through any residues into the soil underneath would provide the necessary earth connection.

## **5.7 Conclusions**

My objectives in this research were to determine whether an ultra-low electric weeding system, suitable for integration into an autonomous weeder suitable for a regenerative agriculture cropping system, could apply a threshold “dose” of voltage and energy to plants growing in the ground outdoors (field treatment) that would achieve more than 90% mortality. I achieved my weed control and energy efficiency targets when treating seedlings growing in the ground outdoors and have shown that the flat plate system tested is suitable for field use as a manual or robotic weeding system in regeneratively farmed cropping fields.

I sought to compare energy expenditure with my earlier studies, and the relative responses for seedlings of two different broadleaf weed species and a grass. In contrast to the suggestions in the meagre literature available, my paired comparisons found that more energy does not appear necessary for effective treatment of in-ground plants than for plants grown and treated in bags. Given equivalent conditions, plants grown outdoors or in a greenhouse have similar responses. Increasing soil moisture increased total energy discharge but was not found to be a significant predictor of increased plant death. Increasing the distance between the earthing and application electrodes reduced energy

discharge, most probably due to increased soil resistance, which may leave less energy available to impact the target weeds.

# **Chapter 6. Modes of Action**

## **EFFECTS OF HIGH INTENSITY MICROSHOCK TREATMENTS APPLIED TO**

### **SMALL WEEDS**

#### **6.1 Publication**

This chapter has not been published.

The extremely low energy demand for effectively killing weeds with PMS raises the question, “But how?”. My observations when applying pulsed microshocks (PMS) to a range of species, pointed to several possible modes of action. Treatments of sufficient field strength caused loss of cell turgor and stem collapse, but the process could sometimes be reversed, in which case turgor returned and the plant was able to recover. If the cell membranes were destroyed by bursting or cell contents were cooked, they could not recover, especially not overnight as was often the case. Something else must be involved. This chapter reviews literature of very short, very high voltage pulsed electric fields (PEF) which are in essence pulsed microshocks (PMS).

## 6.2 Abstract

Observations when applying pulsed microshocks (PMS) to a range of species pointed to several possible modes of action. While high energy electric weeding relies on resistive heating that causes cells to burst or tissues to boil and burn, my calculations suggest the very small energy multiple pulses used in PMS devices are unlikely to cause a significant increase in tissue temperature. Even when hundreds of pulses are applied, the overall energy delivered to the plant seldom exceeds a few Joules and resistive heating can only be minor and PMS might be considered a “non-thermal” method. Treatments of sufficient field strength caused loss of cell turgor and stem collapse, but the process could sometimes be reversed, in which case turgor returned and the plant was able to recover. If the cell membranes were destroyed by bursting or cell contents were cooked, they could not recover, especially not overnight as was often the case. One mechanism that can explain the observed rapid stem wilting and recovery is temporary membrane permeabilisation or reversible electroporation. When the electrical field strength and duration of exposure reach a threshold level, the cell membrane changes and pores develop that allow the intracellular fluids to enter the intercellular space thereby increasing the intercellular fluid’s ionic concentration. This change is measurable as reduced electrical resistance in tissue being treated. My oscilloscope data provide evidence of this phenomenon. Above a critical field strength and duration, the electroporation effect is irreversible and causes damage and death due to cellular component leakages. A secondary symptom was appearance of leaf senescence and progression through to death, possibly via programmed cell death (PCD). I speculate that this could be a reactive oxygen species (ROS) mediated response to drought induced abiotic stress caused by vascular damage brought about by the application of PMS and

while I report that two simple trials did not find expected markers for PCD, I recommend PCD not be discounted.

**Keywords:** electric weeding; mode of action; pulsed microshocks; pulsed electric fields; electroporation; membrane porosity; programmed cell death; abiotic stress

### **6.3 Introduction**

The extremely low energy demand for effectively killing weeds with PMS raises the question, “But how?”. My observations when applying pulsed microshocks (PMS) to a range of species, pointed to several possible modes of action. Treatments of sufficient field strength caused loss of cell turgor and stem collapse, but the process could sometimes be reversed, in which case turgor returned and the plant was able to recover. If the cell membranes were destroyed by bursting or cell contents were cooked, they could not recover, especially not overnight as was often the case. Something else must be involved. What happens when they are treated, and why do they take a long time to die?

This chapter draws together observations of plants’ responses to pulsed microshock treatments with high voltage electricity and reported research in plant science and the parallel disciplines of food processing and extraction and medical treatment, notably for drug therapy. Particular attention is paid to pulsed electric fields (PEF), membrane permeation and electroporation, and programmed cell death. Close similarities found between my experimental observations and reports in the literature provide the base for speculation, and future research that may confirm or reject the proposed mechanisms is identified.

Understanding of pulsed microshocks (PMS), or high intensity pulsed electric fields (PEF) as they are also known, for weeding is lacking in many areas including factors needed for and mechanisms of plant destruction, the modes and safety of electrical equipment, and optimum forms of electrodes (Judaev & Brenina, 2008). The safety of the electrical equipment that I have used was addressed in Chapter 2 “Building a case” (Bloomer et al., 2023b). My studies considered different forms for contact electrode

systems, having used both point positive treatment electrodes as described in Chapter 3 (Bloomer et al., 2022) and plate positive treatment electrodes of different sizes as reported in Chapter 4 (Bloomer et al., 2023a), and probe and plate negative earthing electrodes as described in Chapter 5 (Bloomer et al., 2024).

My research did not focus on proving the actual cause of death when weeds were exposed to short bursts of electricity at high voltages. But my observations during many trials applying PMS to a range of species pointed to several possible modes of action, which are discussed here with reference to published research on the topic. My observations included rapid loss of turgor in plant stems (regularly followed by slow recovery of turgor), exudation of sap droplets following treatment of broadleaf weeds, distortion of developing grass leaves, shrivelling of stems, leaf chlorosis and wilting in mature leaves, and the collapse and death of plants. Notably, symptoms could be very slow to express, sometimes taking up to a week before any effect of treatment was visible and several weeks for plants to die. Plants that seemed affected did not always die. Deducing the likely mode of action of PMS will better allow prediction of factors that influence effectiveness and may enable improved treatments.

## **6.4 Mechanisms of destruction**

Perhaps the oldest methods of weed killing are pulling plants out of the ground and leaving them to desiccate or cutting or breaking them to separate the roots from the shoots. Severing a plant at the hypocotyl will separate roots and shoots and the plant will die (Merfield, 2023). Some of my treatments caused death of the hypocotyl region, effectively separating roots from shoots, and plants subsequently died. However plants,

plant cells, and plant parts can also be killed or triggered to self-destruct in response to other applied stresses.

#### **6.4.1 Lethal temperatures**

Videos of commercially available high-voltage weeding in operation often claim immediate effect (The Weed Zapper, 2018) although others note that several days pass before plants are obviously dead or require re-treatment (Yunker Farms, 2022). The mechanism of action of such high energy treatment is generally attributed to resistive heating (Diprose & Benson, 1984), the plant's electrical resistance causing heating of tissues as the current is applied, in the same way that an electric bar heater works. The result is rapid heating of cell contents to above boiling point and subsequent rupturing of cells. A contrary suggestion is that a shockwave, a pressure buildup caused by the electrical impact (Vasilenko & Sakalo, 1971; Svitalka, 1976), causes cell disruption, although this is also refuted (Baev & Savchuck, 1974).

However cell death can occur well below boiling point, depending on species and other factors. In a study of *Glycine max* L. (soybean) and *Elodea canadensis* Michx (elodea), tissue temperatures over 52°C killed more than 40% of cells immediately after treatment (Daniell et al., 1969). At 57°C, more than 90% of soybean leaf cells had disintegrated chloroplasts and vacuole collapse. The energy required to elevate tissue temperature depends on the mass of material, its specific heat capacity and the change in temperature required. It can be calculated using Equation 6-1, the specific heat capacity formula (Szyk, 2024).

$$E = m \times \Delta T \times c$$

Equation 6-1

Where E is energy (J), m = mass (g),  $\Delta T$  = change in temperature ( $^{\circ}\text{C}$ ) and c = specific heat capacity ( $\text{J}/(\text{g } ^{\circ}\text{C})$ )

The specific heat capacity of plant sap varies. For fresh water it is about  $4.18 \text{ J g}^{-1} \text{ } ^{\circ}\text{C}$  and for salt water is slightly lower. Small *Amaranthus powellii* (redroot) plants such as I have used in trials have a fresh above ground mass of between 2.5 and 5 g, but only a section of stem (the hypocotyl) needs to be killed to destroy the plant. Assuming a specific heat capacity of fresh plant tissue similar to water of  $4.18 \text{ J (g } ^{\circ}\text{C)}^{-1}$ , and a fresh plant density of  $1 \text{ g cm}^{-3}$ , raising the temperature of a 1 g section of plant stem from summer ambient temperature of about  $27 \text{ } ^{\circ}\text{C}$  to a probable-death temperature of  $57 \text{ } ^{\circ}\text{C}$  would require about 125 J of heat energy, which is over 30 times more than the energy discharged in a typical lethal dose using my equipment. Something else is involved!

## 6.4.2 Programmed cell death

Literature reviews identified programmed cell death (PCD) as a possible plant response to various stressors and as a cause of death in plants. Plant PCD is a natural phenomenon, and appears to include chloroplast influences, perhaps by generating reactive oxygen species (ROS) which are produced mainly in the chloroplast, mitochondria and peroxisomes but also in other organelles including the cell membrane and cell wall (Das & Roychoudhury, 2014). Different types of plant PCD have been identified (van Doorn et al., 2011). “Vacuolar” plant cell death involves vacuole membrane (tonoplast) ruptures and can take days after which vacuolar hydrolases rapidly destroy the protoplast and, in some cases, the whole cell and cell wall. The vacuolar cell death process is critical in

tracheary element differentiation for xylem and phloem, with total removal of cell contents essential to create hollow conducting tubes (Kwon et al., 2010). “Necrotic” plant cell death is induced by abiotic stresses and typically involves mitochondrial swelling, rupture of the plasma membrane and loss of intracellular contents. Necrotic plant cell death is faster than vacuolar, taking from minutes to a day. Diagnosis by a combination of electron microscopy and assessment of mitochondrial dysfunction characterised by decreased oxygen consumption, ATP production, and increases in ROS and reactive nitrogen substances is recommended. Hypersensitive response is another plant cell death process, induced by pathogen recognition and abiotic stresses. It is typically like necrosis but often shows tonoplast rupture which requires vacuolar processing enzymes (van Doorn et al., 2011). Mediated by proteases which hydrolyse or degrade proteins, it occurs locally at the primary infection site (Thomas & van der Hoorn, 2018).

Senescence can be controlled by genetic and epigenetic factors responding to external stimuli. Ay et al. (2014) state that the leaf senescence programme is influenced by developmental and stress-related triggers which lead to the induction of senescence-associated processes such as recycling of resources, a decrease in photosynthetic activity, and chlorophyll degradation. Leaf senescence can be triggered by drought stress, and is associated with increasing levels of ROS (Petrov et al., 2015).

A plant or plant part such as a leaf will respond to abiotic stresses in different ways as the result of a complex mix of genetically programmed, age-related developmental stages and stress response pathways (Rankenberg et al., 2021). Abiotic stress resilience is most visible in leaves but can be seen in roots. During early leaf development, leaves are not responsive toward senescence-inducing factors, probably to protect younger leaves.

Studying plant reprogramming of growth in response to mild osmotic stress, Skirycz et al. (2010) observed the “growth was remarkably adaptable” and varied between juvenile and mature leaves. For instance, while ethylene promotes senescence in mature leaves, it does not in young developing leaves, where it instead reduces cell proliferation and expansion (Graham et al., 2012).

Environmental stressors, which include excessive light, mechanical damage, insects, and diseases, can affect photosynthesis, transpiration, and gene expression, and induce numerous other systemic changes. While local action stressors such as these affect plants at different points, a whole-plant response may be required to ensure coordinated physiological responses (Sukhov et al., 2019). Plants’ responses to wounding include activation of calcium and reactive oxygen species (ROS), plant cell death pathways or generation of long-distance signalling via ROS or electric signals (Suzuki & Mittler, 2012). Sukhov et al. (2019) define three main types of long-distance stress signals: hydraulic, chemical, and electrical signals. Electrical signals are further divided into three types, all associated with electric-potential differences on the plasma membrane. Action potential reactions which last for seconds to tens of seconds are associated with “non-damaging irritations” such as electrical currents, cooling, touch and some chemical agents. Variation potential reactions, which Sukhov states are unique to higher plants, last for minutes to tens of minutes, and are associated with damage from burning, crushing and pricking. System potential changes are self-propagated by the plasma membrane, and while stimuli are unclear, it may be associated with H<sub>2</sub>O<sub>2</sub> propagation (Suzuki & Mittler, 2012).

### **6.4.3 Electrical stimuli**

Electrical stimulus can cause membrane breakdown (Weaver & Chizmadzhev, 1996) and short strong electric pulses do kill microorganisms without heat. Svitalka (1976) claimed electric spark treatments with a field strength of  $2.5 \text{ kV cm}^{-1}$  caused flashover that damaged the plant through pressure from a shockwave front. Without flashover, damage was attributed to electro-biochemical and structural changes caused by conductivity currents. Thermal factors, electrical and magnetic field strengths were not significant. However, Baev and Savchuck (1974) had dismissed the shockwave front possibility, identifying that the current flowing through the tissue increased damage with initial discharge energy up to 0.2 J after which flashback occurred. In laboratory testing, a spark channel formed between the electrode and leaf and continued on the surface of the leaf, stem and root, indicating sparking between the root and soil (earth) (Mizuno et al., 1990). Transpiration reduced, indicating that vascular bundles were destroyed. Repeating pulses reduced electrical resistance in the plants, they suggested due to cell wall damage increasing path conductivity. Yudaev (2019) found weed tissue damage, from alternating current (AC) electrical pulses of 10 ms, occurred when cell membranes became fully permeable rather than semi-permeable because pores greater than a critical diameter were created allowing intra- and inter-cellular fluids to mix. This led to loss of cell viability and was characterised by increased conductivity in treated tissues after treatment.

### **6.4.4 Pulsed electric fields**

A comprehensive review of the application of pulsed electric fields in the food industry completed by Aguilar-Machado et al. (2022) showed that the effect of electric fields on plant tissues has been well documented. They reported that it has been subject of patents

from the early twentieth century (Schwerin, 1903), and note the 1947 work of Dedek studying sugar beet tissues that showed a field strength of only  $40 \text{ V cm}^{-1}$  caused a significant increase in electric current during treatment which was suggested to be due to cell damage. Irreversible electroporation is used industrially as a “gentle food technology” using non-thermal PEF (Janositz & Knorr, 2010). It is used in the extraction of cellular substances of value, and has been applied in the areas of sugar extraction from beets, pigments from red wine grape skins, juice from apples, and energy efficient drying of green biomass for biofuel production (Sack et al., 2008; Sack et al., 2010).

For living plants the effects can be variable, increasing or decreasing growth, and appear to be driven by the field strength and duration of exposure. When *Arabidopsis thaliana* seedlings were treated with nanosecond pulsed electric fields and growth was assessed after 7 days, the observed growth stimulation was suggested to be a stress response, possibly to  $\text{H}_2\text{O}_2$  or effects of increased cytosolic calcium leaked from intracellular calcium stores (Eing et al., 2009). At higher energies or pulse lengths the growth increase was overpowered by necrosis from plasma membrane permeabilisation. Lower field strengths of 2 kV ( $5 \text{ kV cm}^{-1}$ ) enhanced growth with either 10 ns or 100 ns pulses. At high field strengths of 20 kV ( $50 \text{ kV cm}^{-1}$ ) all plants died when subjected to 100 ns pulses, whereas 10 ns pulses showed no apparent effect. Moderate field strength treatment at 4 kV and 8 kV ( $10 \text{ kV cm}^{-1}$  and  $20 \text{ kV cm}^{-1}$ ) showed no effect after 7 days. At a set 100 ns pulse, low dose-energy increased growth relative to untreated, but the growth dropped to zero with increasing dose energy, an effect typical of plasma membrane permeabilisation in which pores form and enable cytoplasm to leave the cells which subsequently die. Plant growth increased with 10 ns pulses regardless of treatment energy up to  $4000 \text{ J kg}^{-1}$ . Songnuan and Kirawanich (2012) found *Arabidopsis* growth increased by up to 80% the

second week after treatment with 10 nsPEF at 10 kV cm<sup>-1</sup>. Nian et al. (2013) reported 10 kV cm<sup>-1</sup> nsPEFs could increase *A. thaliana* seedling growth up to an energy level after which treatment was lethal. Su et al. (2021) concluded nitric oxide (NO) is highly likely to be the factor stimulating or suppressing growth depending on its concentration.

#### **6.4.5 Electroporation (electropermeabilisation)**

Living cells have a potential (resting) electrical difference across their membranes of about 150 mV (Ersus & Barrett, 2010). Application of short, high intensity electric pulses alters the transmembrane potential difference in proportion to the cell's radius and the intensity of the applied electric field (Mir et al., 2005). Once the nett of resting and induced potential differences exceeds about 0.2 – 0.4 V (Mir et al., 2005), conductive channels are formed within nanoseconds, and due to pore expansion enabling entry of ionic fluids, the membrane becomes highly permeable after a few microseconds, a process known as electroporation (Weaver & Chizmadzhev, 1996; Eing et al., 2009; Zhang et al., 2014; Thompson et al., 2016; Jiang et al., 2020; Thamkaew & Gómez Galindo, 2020; Shorstkii et al., 2022). Membrane permeability is increased at a voltage of around 0.5 – 1 V across the membrane (Sack et al., 2010) and membranes rupture at a critical value of about 0.7-2.2 V (Angersbach et al., 2000). Below a certain level the process is reversible and the membrane can return to its normal state and the cell will survive, but if exposure is too great, the electroporation effect is permanent and causes irreversible damage and death (Kotnik et al., 2012; Songnuan & Kirawanich, 2012) as a result of cellular component leakages (Haberl et al., 2013).

The electroporation effect can be limited to the cell membrane without impacting organelles by adopting longer, lower voltage pulses of microseconds to milliseconds at

about  $1 \text{ kV cm}^{-1}$  (Haberl et al., 2013). Ersus and Barrett (2010) investigated responses of onion plant tissues to 100 ns PEF and found  $167 \text{ V cm}^{-1}$  increased ion leakage and  $333 \text{ V cm}^{-1}$  caused cell membrane rupture. Comparing the effects of millisecond versus microsecond PEF treatments on the microalga *Chlorella vulgaris* in cultures, Luengo et al. (2015) found that field strengths over  $4 \text{ kV cm}^{-1}$  caused irreversible electroporation using millisecond range treatments but fields strengths over  $10 \text{ kV cm}^{-1}$  were required when microsecond range pulse durations were used. However, the specific energy required was about ten times higher for the longer treatments. Gonzalez and Barrett (2010) reported studies showing that PEF treatments damage plant membranes at ambient temperatures using “moderate electric fields of 0.5 to 5.0 kV/cm within  $10^{-4}$  to  $10^{-2}$  s”. Pulse frequency also affects permeabilisation, which is higher below an apparent threshold of about 1 Hz (Asavasanti et al., 2012). Considering apple, potato and carrot tissues treated by pulsed electric fields, Lebovka et al. (2002) found an optimal field intensity of about  $400 \text{ V cm}^{-1}$  gave lowest power consumption for highest material disintegration. They noted that optimisation requires understanding relationships between plasmolysis, electric field intensity and treatment time, and that conductivity measurements help determine tissue disintegration.

Membrane damage after lower intensity PEFs are applied can be reversed when PEFs are removed. This is evidenced by the decreasing conductivity of plant tissue over a period of at least 24 hours (Lebovka et al., 2001) although the resealing of electropermeabilised pores may be very rapid taking only seconds or a few minutes (Knorr et al., 2002). While resealing may be fast, re-establishing the initial osmotic status of the inter- and intracellular fluids would take longer and would be affected by soil moisture tension and plant transpiration rates.

Used in medical therapy to increase permeability to drugs, electroporation uses very high voltage nanosecond pulses to temporarily increase pore size, allowing drug molecule passage (Songnuan & Kirawanich, 2012). The membrane repairs the pores, returning its integrity. However excessive voltage or pulse duration can irrevocably change pores, leading to cell leakage and death. Songnuan and Kirawanich (2012) presented a boundary diagram in which pulsed electric fields of different pulse lengths and strengths have effects in living cells. Below the lower threshold boundary there is no effect. Above the upper boundary, necrosis occurs because the critical temperature is exceeded.

#### **6.4.5.1 Measurement of effects**

Studies of frost damaged plants showed that the rate of electrolyte leakage varied directly with the extent of tissue damage (Murray et al., 1989) therefore conductivity can be used to assess changes in membrane integrity. There is an initial rapid increase in conductivity associated with the apoplast, followed by a slower second stage that is related to the plasmalemma (Gonzalez & Barrett, 2010). Electrical impedance in biological tissue is complex due to the different responses of different components at different electrical frequencies. Bioimpedance spectroscopy using a range of frequencies allows non-invasive monitoring of biological material. The impedance of intact cell membranes is very high at low frequencies so most current passes through low resistance extra-cellular fluids. As frequency increases, the impedance of the cell membrane decreases and the current passes more easily through it (Bera, 2018). As electrical treatments by PEF cause the membrane to become more porous and there is interchange of inter- and extra-cellular electrolytes, there is an increase in the low frequency conductivity range, but there will be little change in the high frequency ranges (Knorr & Angersbach, 1998). A “disintegration index” was developed, based on the results of conductivity in intact and

permeabilised tissues. Bioimpedance measurements used to track the development of membrane damage as a response to single pulse electrical shocks showed membrane porosity increased within 2 seconds and continued to increase for minutes and hours after treatment (Angersbach et al., 2002).

#### **6.4.5.2 Electroporation equipment**

The principles, application and equipment used for clinical electroporation treatments are well described by Mir et al. (2005) and Rebersek et al. (2014). An example used in clinical electroporation treatments is the Cliniporator device (IGEA SpA, Italy) developed through a European project (QLK-1999-00484) (Mir et al., 2005). These designs are similar to the Zapper multi-pulse microshock weeding system (WedaTech Ltd., Hastings, NZ) used in my research (Figure 6-1). While vastly larger in scale, the electrodes I used were combinations of needles (probes) and flat plates. The additional elements in the WedaTech equipment include the Pico Technology Ltd (Cambridgeshire, UK) high voltage electrode and oscilloscope to enable measurement of individual pulses as they are applied and logging of data for subsequent analysis. In my methodology, the electric field was generated from the treatment electrode through the plant tissue and the surrounding soil to the earthed negative electrode.

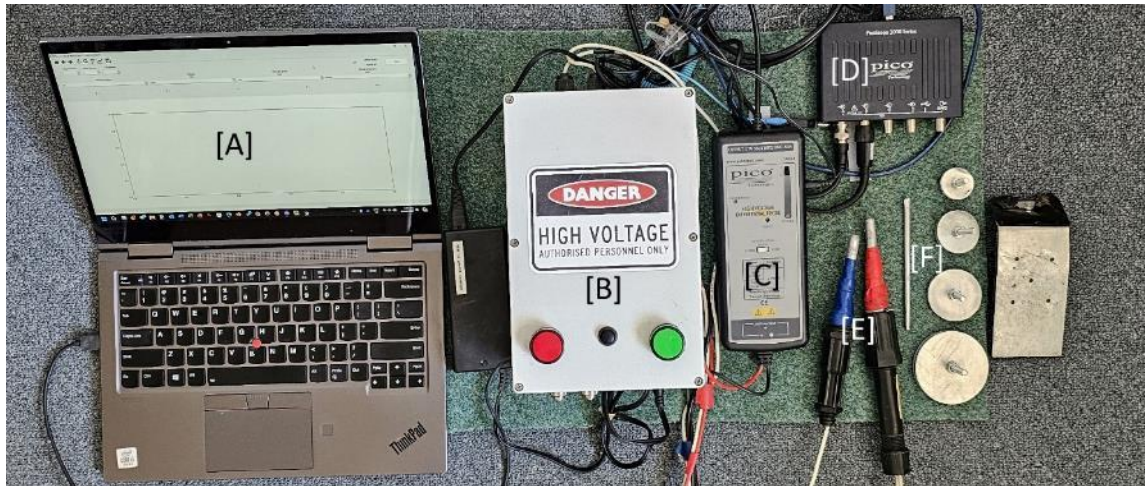


Figure 6-1 Photograph of the WedaTech Zapper multi-pulse system used in my research showing [A] the computer control interface, [B] the high voltage pulse generator, [C] a Pico Technology high voltage electrode and [D] oscilloscope, [E] positive and negative probe electrodes, and [F] alternative needle, circular disc and flat plate electrodes used in my research.

## 6.5 Observations from my research trials

### 6.5.1 Grasses

*Lolium multiflorum* (Italian ryegrass) seedlings were difficult to kill using my equipment. Those with one tiller and two or three leaves had very little biomass, but often survived albeit with reduced final biomass. When I used the original single-spark device to apply 6 kV or 10 kV single pulse shocks to fine *L. multiflorum* leaves, they wilted and collapsed but the plants tended to survive (Figure 6-2). In some cases collapse was instantaneous, much like an electrical fuse blowing when overloaded (Figure 6-2 A), there was a smell of hot grass and steam could be seen as described in Chapter 3 (Bloomer et al., 2022). This showed rapid and destructive heating of the tissues, and simple calculations using fresh mass, the specific heat capacity of water, and energy discharged by the Zapper indicated sufficient heating to elevate the very fine leaves to boiling point.

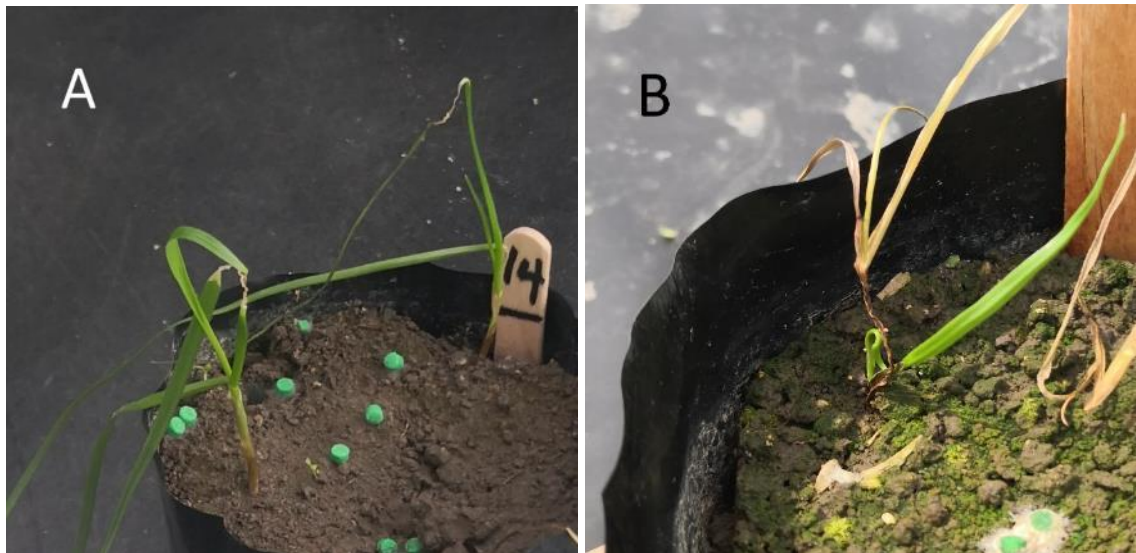


Figure 6-2 Photograph of *Lolium multiflorum* seedlings after treatments showing ([A] - top right) a leaf shrivelled by electrode contact but an otherwise healthy plant, ([A] – left) a plant with leaf tissue distorted in a concertina and ([B] – left) a new tiller emerging from an otherwise dead plant.

In most cases, the larger *L. multiflorum* specimens survived, especially if tillering had commenced, typically by continuing to grow from the leaf base or by generating new tillers from below the soil surface (Figure 6-2 B). This indicated that the single pulse did “blow the fuse” which prevented the energy from reaching or affecting the developing leaves and tillers within the stem base. It was this observation that led to development of multi-pulse equipment, aiming to apply similar total energies but broken into many lower-energy pulses. With the second generation equipment, signs of overheating were absent and plant death rates increased, especially at lower voltages. My later experiments, described in Chapter 5 (Bloomer et al., 2023a), showed that treatment using a flat plate electrode pressing the plants to a dry soil surface could be very effective, even when some tillering had commenced. Despite an assumed direct energy loss to the soil, sufficient must be reaching critical points within the plant.

Lati et al. (2021) measured plant tissue temperatures during treatment with low energy electric shocks and postulated that heat was the driver of plant death. They also noted that grasses are more difficult to kill, and the reduced sensitivity might be due to encircling leaf layers or epidermal waxes or trichomes stopping the electric energy reaching the inner stem or dispersing heat. Thus they retained focus on resistive heating being the main driver causing tissue damage and death. While I did observe excess heating of fine leaves when *L. multiflorum* seedlings were treated with single-pulse shocks, my observations suggest it is the location of the meristematic cells of developing leaves and tillers deep within the plant at or below ground level that protects them as the energy is dissipated and the effective field strength across the membranes is greatly reduced. When applying multi-pulse PMS to plants pressed to soil by a flat plate electrode, the available voltage is delivered very close to the meristematic tissue and the electric field between the treatment electrode and the earth is very short, so the effective field strength ( $\text{kV cm}^{-1}$ ) is much higher. The method is effective and appears to reach the electrical intensity levels required for electroporation, but whether the plant cells are impacted directly through electric current flowing through the plant or via a force field created between the plate electrode and earth is unclear.

### **6.5.2 In-field flatweeds**

In an early proof-of-concept testing of prototype equipment, I applied ~ 10 kV, 6 J direct current (DC) shocks to in-ground weeds in the field in winter using point electrodes. Spark-pulses were applied to the grass *Poa annua* L. (annual poa) and to flatweeds with a rosette growth form including *Capsella bursa-pastoris* L. (shepherd's purse), *Sonchus*

*oleraceus* L. (sow thistle) and *Lepidium didymum* L. (twin cress) in transects across a fallowed vegetable bed (Figure 6-3).



Figure 6-3 Photograph of a transect across a fallowed bed showing weeds treated with pulsed DC electric shocks. Treated plants are marked by toothpicks.

Untreated plants on either side of the transect continued to grow strongly. No symptoms appeared on treated plants for about six days, after which leaf senescence developed, first as silvering of green tissue, then yellowing (Figure 6-4).



Figure 6-4 Photograph of a transect of weeds treated with pulsed DC electric shocks taken 7 days after treatment showing plants yellowing as they develop leaf chlorosis.

Over coming days and weeks, senescence continued to develop with leaves becoming chlorotic and wilting, then collapsing until eventually most flatweeds and some grasses died (Figure 6-5).

Some larger flatweeds regrew from their centres (Figure 6-6) apparently from buds surrounding the stem.



Figure 6-5 Photograph of a transect across a fallowed bed 26 days after plants were treated with pulsed DC electric shocks. Treated plants are marked by toothpicks, and some have completely disappeared. Untreated plants either side of the transect have grown strongly and fully cover the ground.



Figure 6-6 Close-up photograph of a transect across a fallowed bed 26 days after treatment showing dead, senescing, and healthy weeds.

The grasses appeared harder to terminate and most survived, possibly because not all individual tillers were treated or because tillers and leaf meristems were protected from electrocution by their location in the soil. Later trials applying multiple pulse treatments using a flat plate electrode to *L. didymum* found all treated plants grown in the soil outside died, but that it was almost four weeks before definite results were determined

(Chapter 5). The symptoms observed were similar to the first trial, with plants following a normal senescence process of turning yellow and then wilting and dying (Figure 6-7).



Day 0



Day 1



Day 6



Day 12

Figure 6-7 Four photographs taken at intervals of the same *Lepidium didymum* plant treated with a flat disc pressing the plant to soil at time of treatment and at 1, 6 and 12 days after treatment was applied, showing the development of chlorosis and eventual wilting, browning and death.

### 6.5.3 Broadleaf weeds

In my early greenhouse trials using the single pulse Zapper, small *Chenopodium album* (fathen) plants treated by a probe electrode, either sparking to or connected with the stem, collapsed within about a minute to lie prostrate (Figure 6-8). In about 20% of cases, plants recovered overnight (Figure 6-9). Early recovery was typically a strong indicator of eventual survival (Figure 6-10 and Figure 6-11).



Figure 6-8 *Chenopodium album* plants immediately after treatment on 20 April showing stems have collapsed and plants are lying prostrate.



Figure 6-9 *Chenopodium album* plants 1 day after treatment showing two plants lying prostrate and one standing having recovered turgor.



Figure 6-10 *Chenopodium album* plants 8 days after treatment showing one healthy plant and two with collapsed stems and leaves with chlorosis.



Figure 6-11 *Chenopodium album* plants 28 days after treatment. The plant that recovered early is healthy and growing well, the other two plants have died.

The observed collapse and recovery are consistent with the descriptions of electroporation at a level causing electro-permeabilisation of the membranes before allowing recovery and rebuilding intracellular osmotic pressure. I surmise that those plants that did not recover, experienced field strengths sufficient to cause permanent electroporation. Young *A. powellii* plants also collapsed, although older plants were more likely to stay erect, possibly because they have a higher content of lignified tissue. A few days to a week after sufficient doses were applied, treated plants showed strong symptoms of leaf senescence (Figure 6-12) which progressed through to plant death. Leaf symptom expression tended to be associated with observable stem damage (Figure 6-13).



Figure 6-12 *Amaranthus powellii* leaf several days after treatment showing development of chlorosis beginning in the interveinal regions.



Figure 6-13 *Amaranthus powellii* leaves showing red stress symptoms and a collapsed and browning section of stem from treatment point to ground level.

The senescence symptoms observed with *A. powellii* were similar to those exhibited by plants treated with certain herbicides and engendered significant interest among the Herbicide Resistance Management research team members. In the later multi-pulse flat plate trial, three quarters of treated plants had narrowed or shrivelled stems 2 DAT. Some

showed signs of recovery, but by 15 DAT, most that experienced higher field strengths had died. At 2 DAT about 60% of plants receiving the lowest dose showed signs of stem bending but only 20% showed stem narrowing. One third of plants with stem damage and all those with no damage at 2 DAT survived.

The multi-pulse treatments were monitored by oscilloscope (Figure 6-14).

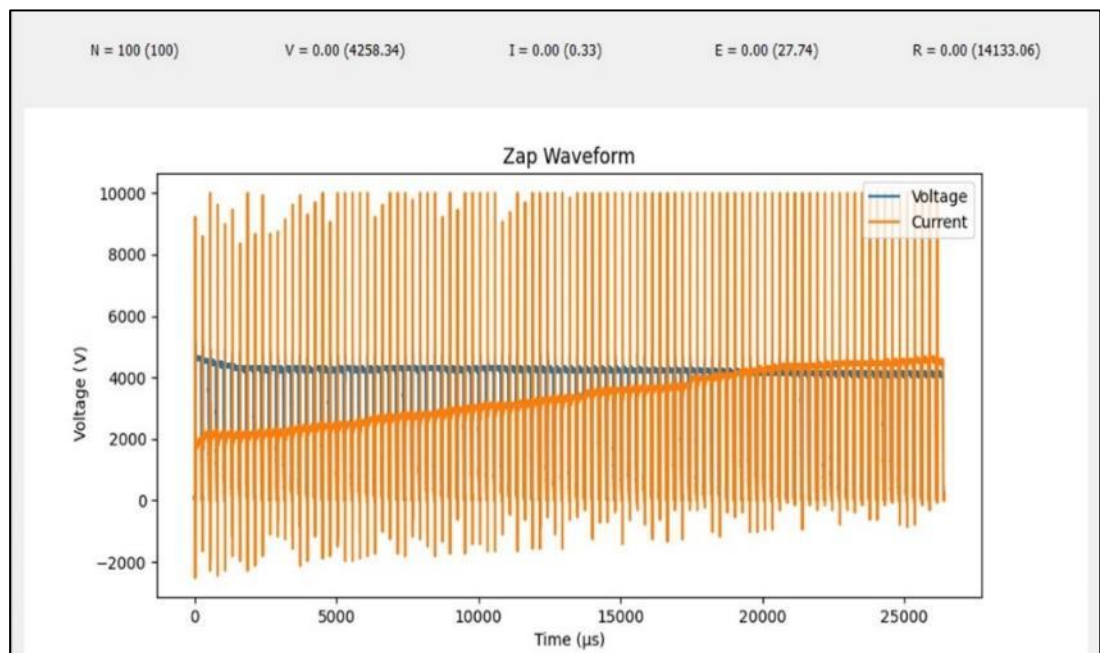


Figure 6-14 Screenshot of the WedaTech Zapper control interface showing a chart of the resulting oscilloscope measurements of voltage and current during application of a treatment to the upper leaves of an *A. powellii* plant. N = logged number of pulses, V = mean voltage, I = mean current (amps), E = calculated energy (Joules), and R = calculated mean resistance (Ohms).

As discussed in Chapter 5, the reducing voltage and increasing current presented in Figure 6-14 must be the result of decreasing resistance in the circuit (Equation 5-1) and by default, in the plant tissue being treated. The pattern is consistent with the work of Mir et al. (2005) and Haberl et al. (2013) and others outlined above describing the rapid development of pores and onset of intracellular fluids moving to the intercellular space as the result of membrane permeabilisation.

Further evidence is that when *Capsicum annuum* (chilli) plants were treated with direct stem contact PMS, they became prostrate within about a minute, and beads of sap were exuded from the stem (Figure 6-15). Note that the stems did not collapse instantly, but gradually sank at an easily observable rate. After about an hour, the stem section from probe contact point to the soil had darkened and exhibited signs of “bruising” within the stem (Figure 6-16).



Figure 6-15 Young *Capsicum annuum* plant treated with stem contact PEF showing beads of sap exudate.



Figure 6-16 Young *Capsicum annuum* plant stem showing tissue bruising under the epidermis.

The appearance of these droplets following treatment with PEF was reminiscent of exudation after cell penetration by aphid stylets, such as is described by Zimmermann et al. (2019).

After about ten days, the treated section of stems had shrunk and turned dark brown, and leaves were wilted and chlorotic (Figure 6-17).



Figure 6-17 *Capsicum annuum* plant two weeks after treatment showing dead stem section and wilted leaves.

Older *C. annuum* plants also exuded beads of sap when treated with PMS even when tissue was becoming woody (Figure 6-18). The affected tissue developed bruising and darkened (Figure 6-19). The same internal bruising symptoms were expressed (Figure 6-20), followed about a week later by development of leaf chlorosis (Figure 6-21) before eventual plant death.



Figure 6-18 Older *Capsicum annuum* plant treated with direct stem contact PEF showing beading sap exudate.



Figure 6-19 Older woody *Capsicum annuum* plant treated with PEF showing beads of sap exudate.



Figure 6-20 Older *Capsicum annuum* plant treated with PEF showing shrinkage at the point of electrode contact (arrow) and internal “bruising” below.



Figure 6-21 Older *Capsicum annuum* plant treated with PEF showing development of chlorosis in leaves.

Moderate strength treatments applied to the lower stem by placing the application electrode 5 cm above the soil surface, and the earth electrode in the soil, successfully damaged the hypocotyl and the plants died. When the same treatment was applied to the top of the stem by placing the application electrode at the top and the “earthing” electrode 5 cm below it, the same stem bruising and shrinkage was observed, and the top section died but the rest of the plant continued to grow normally. This indicates no whole-of-plant response occurred. The observations of *C. annuum* exposed to high strength electric fields tend to support the hypothesis that membranes are made more porous as evidenced by the emergence of sap droplets shortly after exposure to high strength electric fields.

#### **6.5.4 Programmed cell death**

Having noted that programmed cell death (PCD) can cause symptoms like those observed when broadleaf weeds are treated with PMS, an investigation seeking to identify marker compounds was undertaken. In January 2022, a series of trials using *C. annuum* var. “Ma Belle”, applied PMS to leaves and to stems at different positions. Leaves from treated plants were collected, freeze-dried in dry ice at Plant and Food Research Ltd (Havelock North campus, NZ) and with support of Lincoln Agritech Ltd. (Lincoln, NZ) sent to Brett Williams, Advance Queensland Research Fellow at Queensland University of Technology (QUT) in Australia for analysis. Unfortunately the samples arrived during a severe weather event when the campus was closed due to severe flooding. When a verbal report was relayed many months later, it stated that no evidence of PCD markers had been found. It was suggested that the samples should have been collected nearer to the time of treatment. In June 2023, a second trial was undertaken at Lincoln, in which I applied a range of PMS treatments to several species including *Brassica oleracea* (cauliflower) and *Viola x wittrockiana* (white pansies) obtained from a local garden centre. Leaf and stem samples were collected 24 hours and 36 hours after treatment and sent to QUT for testing. Again no PCD markers were reported, but I have not been able to clarify the exact test protocol being used or the specific markers being sought.

#### **6.5.5 Plant resilience**

As described, some broadleaf plants survived even if they initially collapsed. Turgor was restored which is consistent with non-permanent electroporation descriptions in the literature. Other plants survived if planted deeply enough to cover the lower stem section, because new shoots sprouted from buds below ground level. This strongly suggests the

electrical energy had been dissipated into the surrounding earth rather than travelling through the plant to the hypocotyl and roots. This was also the case with grasses, where new leaves and tillers could survive inside the stem base.

A third survival strategy exhibited by some plants, particularly those in the *Solanum* genus, was by regrowing stem tissue or generating adventitious roots. While in some cases the plant died before a new connection or root system could be established, these fleshy plants showed sufficient stored energy and ability to survive by bridging new connections across the damaged stem section, or by adventitious roots formed quickly enough to re-establish connection with the soil and generate a completely new root system. This was observed with *Solanum nitidibaccatum* Bitter. (hairy nightshade) (Figure 6-22 and Figure 6-23).



Figure 6-22 Photograph of treated *Solanum nitidibaccatum* plant with new adventitious root buds forming at healthy stem base above the dead hypocotyl.



Figure 6-23 Photograph of treated *Solanum nitidibaccatum* plant with new adventitious roots that had established into the soil.

The same symptoms were seen on *Solanum lycopersicum* L. (tomato) (Figure 6-24 and Figure 6-25) and *Solanum nigrum* L. (black nightshade) (Figure 6-26 and Figure 6-27).



Figure 6-24 Photograph of treated *Solanum lycopersicum* plant with new adventitious root buds forming at healthy stem base above the dead hypocotyl.



Figure 6-25 Photograph of treated *Solanum lycopersicum* plant with dead lower stem but new adventitious root buds forming at healthy stem base.



Figure 6-26 Photograph of a *Solanum nigrum* plant showing a damaged stem base, leaf reddening and crinkling, and new adventitious buds appearing at the base of the healthy stem.



Figure 6-27 Photograph of a *Solanum nigrum* stem showing new adventitious buds developing above the electrically damaged hypocotyl region.

## 6.6 Conclusion

Seeking explanations of why PMS kill plants with vastly less energy than other weeding systems I have reviewed literature pertaining to plant science, plant processing, and specifically to pulsed electric fields (PEF), membrane permeation and electroporation, and programmed cell death, and I have identified congruences with symptoms I observed when treating young plants with pulsed microshocks. From these parallels I propose that five key conclusions may be drawn.

### 6.6.1 Pulsed microshock effects are not “thermal”

I have demonstrated that the use of high intensity PMS can kill small weeds up to at least 15 cm high with vastly less energy than other systems. It is claimed that most electric weeding relies on high energy to generate resistive heating that causes the cells to burst or even causes the tissues to boil and burn. My research found some evidence of high tissue temperatures when very fine *L. multiflorum* seedlings were treated with single pulse high voltage shocks. While no tissue temperature recordings were made, simple calculations suggest the multiple very-small-energy pulses used with second-generation WedaTech devices were unlikely to cause a large increase in tissue temperature of the broadleaf weeds treated. Indeed, Janositz and Knorr (2010) deliberately used the term “non-thermal pulsed electric fields”. Treatments using PMS are low energy because while they create a very high electric field strength, the pulse duration is very short. Individual pulses are extremely low energy but even when hundreds of pulses are applied, the overall energy delivered to the plant seldom exceeds a few Joules. I also maintained a very long pulse period such that treatment pulses were 100 – 200  $\mu\text{s}$  duration, but the gap between was 50,000  $\mu\text{s}$ . Thus the time for cooling or dissipation was typically at least 250 – 500 times more than the time for heating.

### **6.6.2 Electroporation is consistent with trial data and observations**

My observations were that PMS (or PEF) treatments of sufficient energy (high field strength) caused loss of cell turgor and stem collapse, but that the process could sometimes be reversed, in which case turgor returned and the stem and plant were often able to recover. If the cell membranes were destroyed by bursting or cell contents were cooked, they could not recover, especially not rapidly as was occasionally observed, or overnight as was often the case. One mechanism that can meet the criteria of rapid stem wilting and recovery is membrane permeabilisation or reversible electroporation. When the electrical field strength and duration of exposure reach a threshold level, the cell membrane changes and pores develop that allow the intracellular fluids to enter the intercellular space thereby increasing the intercellular fluid's ionic concentration. This change is measurable as reduced electrical resistance in tissue being treated. My oscilloscope data provide evidence of this phenomenon. Above a critical field strength and duration, the electroporation effect is permanent and irreversible and causes damage and death due to cellular component leakages caused by necrosis from plasma membrane permeabilisation.

### **6.6.3 Programmed cell death should not be dismissed as a factor**

While presence of markers of programmed cell death (PCD) was not confirmed in either of the small trials conducted, it should not be discounted. Some, possibly secondary, process stimulates leaf senescence which then follows a typical path to plant death. My trials indicate that plants can take days or weeks to die after application of PEF. Typically there can be a delay of several days before any visible symptoms appear, after which a sequence of senescence and death follows. Various forms of PCD are described and can

take days for completion. Similarly, hypersensitive response can be induced by abiotic stresses. Senescence need not be fast. Environmental stressors can induce numerous other systemic changes and may require whole-plant response. Plants' responses can include activation of plant cell death pathways or generation of long-distance electrical signalling associated with electric potential differences on the plasma membrane.

#### **6.6.4 Potential for resistance**

Assuming that electroporation is the primary mode of action, the risk of resistance developing to PMS might seem low because electroporation is a fundamental electrochemical phenomenon that affects all cells, algal, plant and animal. However as evidenced by reversible electroporation, cells do show some ability to recover especially from lower intensity doses. Some species that I treated showed greater ability to resist or recover from my applied treatments, so response variability is in evidence. Finally, biology constantly shows enormous ability to evolve in the face of any external influences be they chemical, biological, or environmental.

I have shown PMS can be an effective tool for controlling weeds, but it should, like any technology, be used with other methods within an overall integrated weed management programme. Even if individual populations do not become resistant, a shift in the weed spectrum in treated paddocks is likely as the more resistant plants come to dominate.

#### **6.6.5 More research is needed**

More research is required to confirm the cause or causes of plant death following application of high intensity low energy PMS. Electroporation and loss of intra-cellular fluids is indicated as the primary response. Bioimpedance spectroscopy, as described in

Section 6.4.5.1, is recommended as it allows non-invasive monitoring of membrane integrity over time.

Current evidence suggests that the senescence symptoms observed following application of PMS are a secondary response to abiotic stress, possibly the electrical treatment itself, or possibly another stress factor such as drought stress caused when the vascular tissue is disrupted and cannot be repaired. If the senescence response is related to drought induced PCD, it might be evidenced by ROS and could be assessed by monitoring levels of H<sub>2</sub>O<sub>2</sub> or cytosolic calcium.

Better understanding of the mode(s) of action would enable better targeting of treatments, potentially increasing effectiveness and efficiency. Regardless, the suite of trials conducted demonstrate that when applied high intensity low energy PMS cause damage that kills the cells of the hypocotyl, the roots and shoots are separated and unless the gap can be bridged, the plant dies.



# Chapter 7. Concluding Discussion

## LESSONS AND DIRECTIONS FROM 5 YEARS OF ULTRA-LOW ENERGY ELECTRIC WEEDING RESEARCH

Through my research, a new weed management tool has been identified, proven in concept and in practice, and understanding of potential modes of action has been gained. The research undertaken demonstrated successful electric weed treatment using a fraction of the energy of any other weeding system and allowed unique insights into the energy discharged to plants and the near instantaneous response of cell membranes to the application of pulsed microshocks (PMS). I have successfully demonstrated a novel method for controlling escape weeds in an integrated weed management system and point to possible modes of action that justify further research.

### 7.1 Research imperative

The genesis of this research was a review of non-herbicide weed control technologies within the overall Herbicide Resistance Management research programme (MBIE Endeavour Fund C10X1806 – 2019 – 2023). Herbicide resistance management is an increasing issue in New Zealand and globally and alternative weed control techniques are required.

The context for this research was controlling “escape weeds” following a herbicide or mechanical weeding operation in vegetable and arable weed management crops. With carbon accounting increasingly part of business and spikes in energy costs, energy requirement was a key factor for consideration. My work in minimum tillage cropping

and in regenerative vegetable production highlighted the presence of dry crop and cover-crop residues adding further constraints to weeding methods that might be adopted. Rapid advances in mechanical automation and machine vision coupled with artificial intelligence have made autonomous robotic selective weed management possible but a method to treat the identified weeds was needed.

## **7.2 Systems considered**

Abrasion, hot foam/hot water, high energy electrical, and laser weeding systems were reviewed, assessed and dismissed. Abrasion requires a source of grits and has a high energy demand for compressing air, and the system available for me to test did not have satisfactory weed control performance. Hot foam has a very high energy demand, the system I observed used large volumes of water and could not effectively treat weeds between crop plants. Only one electric weeding system was available in Aotearoa New Zealand, and it also had a high energy demand, caused flaming which was unsuitable in high crop residue situations, and was impractical in a vegetable or arable cropping context. Laser weeding is also extremely expensive and energy consuming, the systems available are only suitable for newly emerged seedlings near cotyledon stage and no equipment was available in New Zealand. I therefore rejected all four systems.

### **7.3 System selected**

My literature reviews uncovered 1990s Japanese research and patents for very low energy weeding equipment. That research stopped, patents have lapsed, and the concept appears to have been abandoned. Soviet work into high efficiency high voltage electric weeding is also cited but very little information is available. However the concept of very high voltage of very short electric shocks was intriguing and appeared able to meet the criteria on which my research was to be based.

Goals and objectives were set, and series of trials was conducted, proving the concept of ultra-low energy pulsed microshocks (PMS) to kill small weeds, identifying a practical application method, and demonstrating successful deployment in the field. Careful observations of treated plants identified a range of physical plant reactions that align very well with other research into the effects of PMS on plant tissues and point to new understanding of the mode of action of electric weeding. I postulate that PMS of a certain intensity causes increased cell membrane permeabilisation or electroporation. Opening of membrane pores allows intermingling of intra and extra cellular cell fluids which is detectable using plant conductivity measurements. At low doses the process can autonomously reverse when the treatment ceases and the plant may recover. However a dose of sufficient intensity and duration results in irreversible electroporation and subsequent cell death. If applied in a way that disrupts the hypocotyl and meristems, the outcome is weed death. A secondary effect appears to be involved, in which leaves senesce and the plants subsequently die, possibly due to drought-induced abiotic stress caused when the vascular tissue is disrupted and cannot be repaired.

## 7.4 Research objectives

My research was guided by four key objectives:

1. Define a set of performance criteria for a novel weeding device in response to increasing herbicide resistance, consumer desire for spray-free food, and a move towards regenerative agriculture in crop production.
2. Determine the minimum dose required to kill a range of small broadleaf and ryegrass weeds in a laboratory greenhouse setting.
3. Identify a practical application solution that could be employed to selectively apply such doses to small broadleaf and ryegrass weeds in the field.
4. Review the literature on possible electric weeding modes of action and propose mechanisms that may enable the greatly reduced energy requirements of pulsed multishock treatments.

Seeking a measure by which success might be assessed, I set a goal of successfully killing more than 90% of targeted weeds using less than 2 MJ ha<sup>-1</sup> application energy at 5 plants m<sup>-2</sup>, a plant density considered reasonable for post weeding escapes after treatment by other means (Kaufman & Schaffner, 1982; Vigneault et al., 1990; Coleman et al., 2019).

Is 90% control an adequate target? Clearly 100% control is better, and the more weeds that survive any treatment, the greater the possibility of population increase and of a resistance issue emerging. The 90% target was selected based on Nørremark et al. (2006) who reviewed reported levels of dry matter reduction of annual and perennial monocot and dicot weeds subjected to physical damage in pot experiments.

Application energy of  $< 2 \text{ MJ ha}^{-1}$  is 10% of the energy target suggested by Coleman et al. (2019) for post-weeding escapes, and 5% of the target set by (Nørremark et al., 2006) for autonomous inter-row weeding. Note however, that those authors include the total energy need including that for transporting equipment around a paddock, activating any actuator equipment such as robotic arms, and any navigation, vision and computing power required for successful operation.

## **7.5 Research progression**

A literature review of herbicide resistance, social and regulatory pressures to minimise herbicide use, and of non-herbicide weed control technologies is presented in Chapter 2. In a series of staged research trials, I demonstrated proof of concept in a greenhouse setting, a method for practical application, and effectiveness in a field setting. Chapter 3 describes the use of very carefully positioned aluminium point-probe electrodes to apply very short direct current (DC) electrical pulses at high voltage (pulsed microshocks or PMS). This showed that PMS could kill small grasses, flatweeds and various types of broadleaf weeds. The precision required to reliably place such electrodes is not practical for real world deployment, so Chapter 4 reports on second stage trials that swapped the point-probes for a flat plate pressed to the weeds' leaves and showed that similar results could be obtained in bagged plants growing in a greenhouse. The final trial stage, presented in Chapter 5, took application to a field setting, treating weeds growing in bags and weeds growing in the ground. The plate electrode can be used either pressing against the foliage only or pressing the entire weed to the ground, although the discharged energy is typically higher when ground contact is made. The overall energy required is orders of magnitude lower than other reported weed control technologies, including herbicides.

Chapter 6 draws together my electrical records and observations of plant responses to treatments applied in over 25 trials and compares them with descriptions of plant responses reported in plant science, electric weeding, and food processing literature. I postulate that by utilising very short, very high voltage pulses, the principal mode of action when using PMS is not thermal damage but the result of increased membrane porosity. A secondary effect is indicated in which leaf and then plant senescence is stimulated by drought-induced abiotic stress caused by the vascular disruption. While specific experiments were not conducted, there is considerable alignment between my recorded application data and observations, and the body of published evidence. This area is recommended for further research.

## **7.6 Research outcomes**

### **7.6.1 PMS control is effective and highly energy efficient**

I have demonstrated that the use of PMS (or PEF) can kill weeds up to at least 15 cm high with vastly less energy than other reported systems. Assuming 5 weeds m<sup>-2</sup>, a reasonable number for treating escape weeds following herbicide application with some resistance, full control was achieved for small *L. didymium* and *A. powellii* plants at 25 kJ ha<sup>-1</sup>, and for in-ground *L. multiflorum* at 363 kJ ha<sup>-1</sup> (leaf contact only) and 555kJ ha<sup>-1</sup> (leaves pressed to soil), all well below my initial target of 2 MJ ha<sup>-1</sup>. PMS is orders of magnitude more energy efficient than any other identified weed control technology for controlling small weeds such as escapes following treatment with herbicides.

### **7.6.2 Responses and effectiveness vary between species**

Early success in trials applying PMS to *C. album*, *A. powellii* and *C. bursa-pastoris* were tempered by failure to adequately manage *L. multiflorum*, especially once tillering had commenced. Buried meristems or side shoots reduce effectiveness, apparently by allowing the electric field to moved directly into the soil before damaging the plant cells. Fleshy weeds including several *Solanum* species showed ability to recover from treatments using PMS by bridging damaged tissues with new growth or by generating adventitious roots and creating new root systems. Later trials with a flat plate showed that *Solanum* species can be controlled using PMS, as can *L. didymium*. Even *L. multiflorum* seedlings were adequately controlled in the field, despite tillering having been initiated.

### **7.6.3 PMS can be effectively and selectively deployed in a field setting**

The use of flat plate treatment electrodes either placed on leaves only or pressing plants to the soil surface is a simple way to apply PMS and I showed it can be both energy efficient and an effective way to control weeds. This approach is suitable for deployment in a handheld device treating individual weeds. With further engineering multiple units could be deployed in banks as a boom or strip weeder, and still have a low overall energy requirement. A simple trial demonstrated that non-target seedlings immediately adjacent to treated weeds were not affected by PMS unless physically contacted.

### **7.6.4 PMS could be deployed robotically**

The plate electrode I developed is effective and highly energy efficient and requires less precise positioning than does a needle or probe electrode. Thus PMS could be delivered by an applicator on a robotic arm using, for example, the design adopted by Ecorobotix in their solar powered Generation 1 machine, to treat only the targeted weeds and avoid impacting the crop plants.

### **7.6.5 Effective grounding is essential and readily achieved**

As with any electrical circuit, effective earthing is essential. However an earthing electrode can be relatively small, and I showed that a 5 mm diameter aluminium rod was effective in outdoor applications especially if the underlying soil was moist. High-energy systems pull steel discs through the soil as grounding units, but in lower powered robotic system an earthing probe mounted near the treatment electrode would be satisfactory.

### **7.6.6 Dry soil acts as an effective insulation material**

When applying PMS via a plate electrode pressed to the plant and soil surface, a thin layer of dry soil is sufficient to minimise direct energy loss making a higher proportion of potential discharge travel through the plant tissues and increasing efficiency and effectiveness. The earthing electrode should penetrate the dry soil layer for most effective application.

### **7.6.7 PMS treatments are not thermal controls**

Treatments using PMS are low energy because while they create a very high electric field strength, the pulse duration is very short. Individual pulses are extremely low energy and even when hundreds of pulses are applied, the overall delivered to the plant seldom exceed a few Joules. I also maintained a very long pulse period such that treatment pulses were 100 – 200  $\mu\text{s}$  duration, but the gap between was 50,000  $\mu\text{s}$ . Thus the time for cooling or dissipation was typically at least 250 – 500 times more than the time for heating. Non-thermal effects are indicated.

### **7.6.8 Electroporation is the probable mode of action**

My observations show that PMS treatments of sufficient field strength and duration caused loss of cell turgor and stem collapse, but that the process could sometimes be reversed, in which case turgor returned and the stem and plant were often able to recover. One mechanism that can meet the criteria of rapid stem wilting and recovery is temporary membrane permeabilisation (reversible electroporation). Changes in membrane permeability can be measured using bioimpedance spectroscopy, which is a recommended tool for further research.

### **7.6.9 Avoid the risk of resistance developing**

The postulated mode of action acts at a fundamental electro-physical level on cell membranes and appears consistent across cells in different types of tissues including algae, plants, and animals. While it might seem difficult to see how resistance could evolve to this treatment, biology repeatedly demonstrates enormous ability to evolve and, especially given the variable effectiveness against different species, repeated reliance on PMS as the only treatment could see the weed spectrum in a field shifting, so integration of PMS and alternative control methods within an overall weed management system is recommended.

## **7.7 Opportunities for further research and development**

### **7.7.1 Optimisation trials**

#### **7.7.1.1 More species**

While I have tested 9 species and all are susceptible to electrocution, the energy levels vary. Because computing now allows identification of plants by species, understanding specific effective dose requirements could optimise treatments.

#### **7.7.1.2 Different sized plants**

I tested weeds at an established but relatively young stage, as might typically be found as survivors after an ineffective herbicide treatment. I did not try to determine minimum effective doses for much smaller seedlings, or for larger plants. There is a role for PMS equipment on both these alternative cases and further studies to identify requirements are recommended.

### **7.7.1.3 Different soils and soil moisture levels**

Most of my trials used the same soil type, collected from my workplace in Hastings although some used other local soils. I did not test the PMS treatment on peaty soils, heavy clays, or very sandy soils, all of which have different electrical properties, so I do not have evidence that the same doses will be effective in such situations. I have applied PMS to plants in a wide range of soil moistures and have reported in Chapter 5 (Bloomer et al., 2024) that while soil moisture did affect treatment energy efficiency, it was not a significant variable in effectiveness. I noted that a layer of dry soil at the surface acted as an electrical insulator and reduced energy discharge. More work could be done to identify optimum and fringe conditions, and any boundary limits to effective deployment.

### **7.7.1.4 Different pulse and voltage combinations**

Optimisation requires understanding relationships between plasmolysis, electric field intensity and treatment time, and may be aided by conductivity measurements to help determine tissue disintegration. I have used many combinations of different voltages, pulse lengths, pulse number and pulse period but the electrical current is determined by the design and components of the equipment at my disposal. Similarly, I used a relatively long pulse period because of limits in the capacitors' recharging speed in my equipment. For field deployment, a shorter period (higher pulse frequency) would allow faster treatment and increase real work rates. Testing a vastly shortened pulse period should be a priority.

#### **7.7.1.5 Earth electrode designs for commercial deployment**

I have applied treatments to each plant manually, taking some care to ensure the system was well earthed, and checking the earthing probe after each application. The distance between the earth electrode and treated plants has, for most of my trials, been relatively small although the initial flatweed testing had up to 70 cm separation with no apparent reduction in effectiveness. In the field, especially using mechanical and autonomous methods, earthing must be reliable both for effective treatment and for efficient working. In the case of weeding in regenerative or conservation agriculture systems with high surface residue levels, if the probe does not make good soil contact or gets clogged by debris, the system will have poor performance. Research and development need to find suitable earthing electrode design options. Further investigation into how far the earth electrode can be from treated plants and testing alternative earthing methods to find reliable field-ready options is also needed to inform equipment design and operating constraints.

#### **7.7.2 Clarification of mode(s) of action**

While I have identified mechanisms that appear able to explain the symptoms I observed when plants were treated using PMS, I did not conduct specific experiments designed to confirm or reject my postulated modes of action. Bioimpedance spectroscopy is a recommended method for assessing cell membrane permeability/porosity during and after treatments are applied. This would help determine if the hypothesis that electroporation and loss of intra-cellular fluids is the primary response and cause of cell and tissue death is correct. Analysing plants for the presence and levels of ROS including H<sub>2</sub>O<sub>2</sub> or cytosolic calcium could help support the postulation that subsequent senescence is the result of drought induced PCD.

### **7.7.3 Equipment limitations**

The equipment I commissioned from WedaTech Ltd. was designed and built to my specifications especially for this research. The first generation equipment applied single pulse shocks at a predetermined voltage. Its key advantages were that it had relatively simple circuitry, it was very light and easy to carry and use, and it ran independently powered by a small lithium battery. The single spark could arc if there was limited connection to the plant, which allowed effective transfer of energy. While effective in early trials with broad leaf and flat weeds, it was not very effective against grass weeds.

The second generation equipment introduced multiple pulse options, allowed me to control doses carefully and precisely by setting the voltage, pulse length, pulse duration and pulse period, and to simultaneously capture and log accurate data for each pulse applied through a connected computer and interface. The equipment reduced the energy of individual pulses and helped reduce problems with fine grass leaves “blowing the fuse” and protecting the rest of the plant. I was able to attach any type of electrode and use the equipment in a laboratory or in a field if a 230 V power supply was available. It is not suitable for commercial use.

### **7.7.4 Engineering opportunities**

I believe there are commercial opportunities for both Generation 1 and Generation 2 WedaTech style devices. In both cases, safety will be a consideration. I have outlined in Chapter 4 that the equipment I propose has energy and voltage levels similar to an electric fence unit and is thus relatively safe. Compared to some herbicides, many mechanical options, lasers, and the very-high-power electric weeding machines in use, it could be

considered very safe. If necessary, commercial adoption could be accompanied by a brief training session.

#### **7.7.4.1 Gen 1 Zapper**

Robust and simple to operate and charge, a single pulse, light-weight Zapper is an option for a less expensive handheld device used to manually treat weeds at low weed densities. I showed that the prototype equipment could control small flatweeds and broadleaf weeds such as *Chenopodium* and *Amaranthus* when used as a needle probe “spark” applicator. The same unit with a circular disc pressing a plant against the soil surface will probably severely limit the growth of small grasses, perhaps killing them, although I have not reliably tested that in the field. My current mobile phone battery is rated 14.55Wh, which equates to 53.28 kJ, so assuming an average of 5 J pulse<sup>-1</sup>, such a battery would power more than 10,000 pulses, enough for about three hours of continuous operation at 1 pulse second<sup>-1</sup>. A common power tool battery (18V 6.0 Ah) holds over 440 kJ so would last a full workday in any realistic operating context.

#### **7.7.4.2 Gen 2 Zapper**

The second generation multiple pulse equipment could be adapted to work as a handheld device, perhaps with preset doses or with a simply adjusted “dose setter”. The energy discharge required for lethal doses was found to be in a similar range to the Gen 1 system, but the two systems were not directly tested together to compare performance.

An alternative application for the multiple pulse Generation 2 equipment is as a strip-weeder, with pulse voltage and pulse length able to be set, and pulse period adjusted to best match weed density and machine ground speed. A simple estimate shows that using

a 200 mm long plate travelling at  $5 \text{ km h}^{-1}$  the electrode would pass a point in 0.278 s. To apply 100 pulses in that time, the pulse frequency would be  $360 \text{ pulse s}^{-1}$  or 360 Hz, and the pulse period (the pulse length plus the gap between pulses) would be  $2,778 \text{ }\mu\text{s}$ . My trials used pulses in a range of  $25 - 200 \text{ }\mu\text{s}$ , but I used a period of  $10,000 - 50,000 \text{ }\mu\text{s}$  to ensure the machine's capacitors recharged fully between pulses. Higher-quality, higher-power components would ensure rapid capacitor recharge and I believe, but could not reliably test, that a shorter period would have the same effect on the plants. Running at  $200 \text{ }\mu\text{s}$  pulses and  $2,800 \text{ }\mu\text{s}$  period still allows more than 10 times more energy dissipation time than energy application time, so heating is unlikely.

It would be interesting to try adapting an electronic car ignition system to this application. These systems are very robust and reliable. They typically generate  $10 - 30 \text{ kV}$  sparks thus the output voltage is at least as high as used in my research. As an example, an 8 cylinder 4-stroke engine running at 6,000 rpm requires  $400 \text{ sparks s}^{-1}$  so the pulse frequency generated would meet the 360 Hz target suggested above. The first step would be to determine if these systems are open to control for a third-party application.

#### **7.7.4.3 Robotic deployment**

Robotic deployment of PMS is feasible as described in Chapter 1.1.5. A solar-powered autonomous spot-weeder based on the DELTA design could, like the Ecorobotix Generation 1 machine, identify, locate and apply treatments. Advances in artificial intelligence and image analytics can assess size and are beginning to recognise individual species and could be used to apply bespoke doses accordingly. Using energy-efficient DC PMS applied by a plate electrode, effective weed control could be achieved. I note however that Ecorobotix abandoned the Generation 1 design in favour of a larger, tractor

mounted precision sprayer, which I saw in operation at FIRA Salinas in California in September 2023. Like many other companies commercialising robotic weed control systems, they encountered markets preferring to retain human driven machines and to leave the robot as “smart” equipment mounted on a conventional tractor. I believe this will change in time as labour supplies continue to dwindle and become increasingly expensive, and farmers become more familiar with autonomous equipment routinely handling their dull, dirty, and dangerous tasks.

## 7.8 Conclusion

My research objectives were achieved. This research used custom-built high-intensity PMS equipment to test an ultra-low energy electric weeding concept and demonstrated that control of small weeds can be achieved using a fraction of the energy of any other system. Together with careful observations, the detailed pulse data provide strong evidence that the mode of action when plants are exposed to PMS is membrane electroporation. A practical field application method was developed and tested and shown to meet the target goals set.

My goal of successfully killing more than 90% of targeted weeds using less than 2 MJ ha<sup>-1</sup> application energy at 5 plants m<sup>-2</sup>, was easily met. Pulsed electric microshock weed control is a novel ultra-low energy method for use within an integrated weed management system. Combined with automation technologies and artificial intelligence, it offers an autonomous, low-energy, non-chemical, selective weeding system that has broad application, including vegetable and arable crops, orchards and vineyards, amenity horticulture and even roadsides. Further research is required to understand effectiveness and energy efficiency with other species and with plants of other maturities and sizes, in different soil contexts, and to confirm the mode or modes of action of PMS weeding. The method appears suitable for development as a commercial technology for deployment by hand or machine, or as part of an autonomous robotic weed control system.



## Chapter 8. References

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# **Appendix 1 Statements of Contribution**

Chapters 2 – 5 or reproductions of papers published between 2022 and 2024.

The following sections are copies of signed statements made by

the **Candidate**

**Daniel James BLOOMER**

and the **Primary Supervisor**

**Associate Professor Dr Kerry C HARRINGTON**



## Statement of Contribution Chapter 2

DRC 16



GRADUATE  
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### STATEMENT OF CONTRIBUTION DOCTORATE WITH PUBLICATIONS/MANUSCRIPTS

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GRADUATE RESEARCH SCHOOL

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