Project title:	"eyeSpot" – leaf specific herbicide applicator for weed control in field vegetables
Project number:	CP 134
Project leader:	Alistair Murdoch, University of Reading
Report:	**Final Report, July 2020**
Previous report:	**Annual Report, June 2018**
Key staff:	University of Reading: Professor Alistair Murdoch (PI), Dr Nikolaos Koukiasas (Researcher and PhD student), Paul de la Warr (software engineer). Concurrent Solutions llc: Dr Robert Pilgrim, Shane Sanford, Steve Bruekner.
Location of project:	University of Reading School of Agriculture, Policy and Development: (a) Agriculture Building, Earley Gate, (b) Controlled Environment Laboratory, Whiteknights and (c) Crop Research Unit, Sonning Farm. Concurrent Solutions llc (USA): (a) Benton, Kentucky, (b) Nashville, Tennessee and (c) Dallas, Texas (including linked activities not funded by AHDB).
Industry Representative:	Mr Phil Lilley, Hammond Produce Ltd., New Farm, Redhill, Nottingham NG5 8PB.
Date project commenced:	1 April 2015.
Completion date	31 January 2020.

Co-funders	Co-funders of PhD student: Douglas Bomford Trust, Edith Mary Gayton Trust Fund and the University of Reading Graduate School, University of Reading, School of Agriculture, Policy and Development) Co-funder of project: Concurrent Solutions llc funded associated development work, the prototype autonomous platform (robot) and most of the software. Concurrent Solutions llc. and Soya Bean Growers Association jointly funded the glasshouse facility at Benton, Kentucky.

Acknowledgements:

Special thanks are due to technical staff in the School of Agriculture, University of Reading, without whom much of the research would not have been possible. These include, Liam Doherty, George Gibbins and Val Jasper on the Whiteknights campus, and Richard Casebow, Caroline Hadley and David Mclay at Sonning Farm. We also gratefully acknowledge the help of Jennifer Wickens and Victoria Wickens, who were employed as field assistants in 2017 on the project. We also thank three students who participated in the work: two MSc students - successfully completed their dissertations – both going on to PhD programmes elsewhere, and Miss Fern Price-Jones, who was funded by the University to work on the project during summer 2016 as part of Reading's Undergraduate Research Opportunities programmes.

Thanks are also due to Mrs Amy Sanford and Miss Leia Sanford from the USA, who helped in summer 2019 to get the prototype autonomous platform operating in time for the Open Day on 25th July. We are also very grateful to Mrs Amy Sanford for legal advice on IP issues throughout the project. Thanks are especially due to Mrs Mary-Beth Pilgrim for the generous hospitality offered during project activities in Benton, USA during summer 2017.

Thanks are also due to Brian Knight of Knight Farm Machinery for his active interest in the project from its inception and, in particular, its commercialisation potential in the UK.

Finally, we gratefully acknowledge the administrative help from staff at Reading and the financial support and interest of AHDB and all our co-funders – mentioned above.

DISCLAIMER

AUTHENTICATION

We declare that this work was done under our supervision according to the procedures described herein and that the report represents a true and accurate record of the results obtained.

Alistair John Murdoch

Professor of Weed Science

University of Reading

Signature .... Date ....

[Name]

[Position]

[Organisation]

Signature .... Date ....

Report authorised by:

[Name]

[Position]

[Organisation]

Signature .... Date ....

[Name]

[Position]

[Organisation]

Signature .... Date ....

CONTENTS

GROWER SUMMARY

Headline

- Weeds up to the 4-leaf stage could be controlled leaf-specifically by applying one droplet containing either 32 µg of glyphosate or 28 µg of glufosinate-ammonium to a single leaf of the seedling.
- The economic analysis predicted that weed control using plant-specific droplet applications to weeds in UK cabbage crops and leaf-specific applications in UK leek crops would at least maintain and in many cases increase their profitability for growers. These economic benefits include the full estimated costs of an automated system for droplet application. More specifically:
    - In cabbages, three plant-specific droplet treatments with glyphosate droplets resulted in gross margins above the total costs of weed control estimated at £32,000 ha^{-1} for savoy cabbages in 2016, which was significantly higher than the £22,000 ha^{-1} when weeds were managed by conventionally-sprayed pre-emergence pendamethalin. For the white cabbage crops grown in 2017, differences between weed control regimes were not significant largely because the crop competed effectively against the weeds. Importantly however, plant-specific weed control did not reduce the profitability of the crop.
    - In leeks, ten leaf-specific applications of glyphosate droplets at approximately weekly intervals in both 2017 and 2018 and a similar treatment using glufosinate-ammonium in 2018 achieved gross margins of £29-32,000 ha^{-1}, much higher than the £10-17,000 ha^{-1} for the conventionally-applied pre-emergence pendamethalin spray.
    - To support growers in deciding whether to adopt leaf-specific weed control, simulation modelling predicted that there was over an 80% chance that a grower would make more profit by controlling weeds in leeks leaf-specifically compared to conventional spraying. Gross margins after accounting for all weed control costs, were predicted to ***increase*** by more than £10,000 ha^{-1} (per year) in 60% of cases.

- In terms of the efficacy of weed control, over 90% weed control was achieved with leaf-specific droplet treatments, even though herbicide inputs were reduced by up to 82 and 94% in transplanted leeks and cabbages, respectively.
- A prototype platform (Figure 1) for leaf-specific weed control was developed for the project by Concurrent Solutions llc and was demonstrated to growers and other interested parties in July 2019. Commercialisation under present market conditions requires further investment and also a larger market than exists in the UK alone.

Figure 1 Prototype robotic platform specially designed, developed and built for the eyeSpot project by Concurrent Solutions llc in the USA. The platform was designed to treat four rows of vegetables planted in beds of two metres width. The platform is shown at Sonning Farm in July 2019 with four rows of cabbages (50 cm between rows; 30 cm between plants in rows.) The black tapes along each row are the drip irrigation system. The platform was demonstrated to growers and others in July 2019 at an AHDB Open Day, which was organised to disseminate and demonstrate the results of the eyeSpot project.

Background

Weeds and their control play a vital role in maintaining vegetable yields and quality and herbicides are a highly efficient method of managing weeds. Herbicides account for 40% of the total amount of pesticides applied by vegetable growers compared to 31 and 24% for fungicides and insecticides, respectively (Garthwaite *et al.*, 2017). However, improper or inappropriate use of herbicides may have adverse effects on human health and the environment. Even though herbicide use is subject to stringent regulations, the EC Regulation No. 1107/2009, the Water Framework Directive (2000/60/EC) and the Sustainable Use Directive (2009/128/EC) are leading to the loss of herbicide actives and make it more difficult for new compounds to gain approval. This predicament is exacerbated for field vegetable growers because they rely on a limited range of older herbicides released in the 1960s and 1970s, which require a lot of funding and effort in order to keep them in the market.

This project offers a paradigm shift for post-emergence weed control in field vegetables. Some use of chemicals is retained, but the focus was to develop a novel engineering solution. The concept was to control individual weeds plant- or leaf-specifically by applying single droplets of a non-selective, systemic herbicide to the unwanted plants. As far as possible, direct herbicide applications to the crop or the soil were to be avoided.

Overall objectives were to:

- minimize herbicide inputs and meet demand for more sustainable crop production, providing an efficient, cost-effective and environmentally-sustainable means of controlling weeds in vegetables;
- eliminate herbicide drift and reduce run-off to the soil, crop and non-target organisms; and
- provide an alternative to conventional spraying for transplanted field vegetable crops where few post-emergence herbicide options are available.

Plant specific weeding by hand is what growers have traditionally done. Individual plants are examined and the unwanted ones are hoed or removed. Even were the labour available and willing to hand-weed crops, the process is unlikely to be cost-effective and the task is dull, difficult, dirty and perhaps even dangerous (the four “Ds” of robotics).

The proposed system also offers advantages over mechanical intra- and inter-row tillage systems. Energy and fuel use are expected to be lower and the absence of soil disturbance means fewer weed seeds will be stimulated to germinate and the likelihood of soil erosion will be lower.

The project was therefore funded to explore the possibility of achieving leaf-specific weed control using an autonomous platform. The project is an alternative to other possible plant specific weed control systems which have been proposed using directed sprays, lasers or electrocution. The former is currently available and the latter two have been investigated but, as of now, appear to have been deemed unsuitable for commercial development. A detailed comparison of the directed spraying option with eyeSpot was carried out prior to this project receiving funding and perhaps the essence of the difference is that the former targets large individual weeds such as potato volunteers, whereas eyeSpot is designed to reach weed seedlings with a leaf area of 1 cm^2, i.e. soon after they emerge at cotyledon or first true leaf growth stages.

Summary

Precision targeting of herbicide droplets to the leaves of weeds involves use of very small droplets (1-2 microlitres) – so that one teaspoonful (5 ml) was enough to treat 2500-5000 individual weeds if one droplet is put on each weed. Nevertheless, the droplets are much larger than those used when spraying conventionally so that there is no risk of spray drift. There is still a potential for spattering on impact and some shattering of droplets on ejection from an applicator and the droplets are likely to be deflected by wind. We therefore carried out preliminary trials with a prototype droplet ejector to investigate how applicator pressure and distance from target affected spattering. The effect of wind on deflection of droplets was also investigated in a multifactorial experiment comprising windspeed and direction, applicator pressure and distance from target as factors. Provided windspeed and direction are known, deflection could be modelled and compensated for. Applicator pressure of 138 kPa (20 psi) avoided all spattering and droplet shattering after ejection in our tests.

Our initial experiments all related to use of glyphosate – in many ways an ideal active ingredient because of its mode of action, efficacy against most weeds, low cost and, most importantly for droplet applications, its systemic behaviour in plants. To reduce the risk of creating a selection pressure for glyphosate resistance in weeds and to explore alternatives should glyphosate lose its approval, we have also tested glufosinate ammonium and 2,4-D and mixtures of these products. Although

glufosinate ammonium has limited systemic action, it achieved reasonable efficacy. 2,4-D is systemic, but would not control grass weeds.

Doses applied in every case are linked approximately to the ground cover of the weeds. As a general recommendation, weeds that are up to the 4-leaf stage can be controlled with a dose of 32 µg of glyphosate and 28 µg of glufosinate-ammonium when these amounts are applied plant-specifically, that is as a single droplet per seedling. There is a potential issue as regards approval, for although the amount of product applied to each square metre of field will always be less than the permitted dose, the same cannot be guaranteed for every square centimetre. There are of course 10000 cm^2 in each square metre but the current approvals were devised for broadcast spraying and do not take account of the focussed targeting of individual plants or leaves achievable by robotic weeders.

In field trials (2016 to 2018) with plant- or leaf-specific weed control droplets, herbicide inputs were reduced by over 90% and 70% in cabbages and leeks, respectively, compared to a pendimethalin pre-emergence spray. Efficacy of weed control and crop yields were not significantly lower than in the hand-weeded, "weed-free" controls.

Financial Benefits

A detailed economic analysis showed that, after accounting for all fixed machinery costs and all the variable costs of weed control, leaf-specific weed control could ***increase*** profits by over £11000 and £1500 per hectare per year for leeks and cabbages, respectively. Bearing in mind that these are average estimates, a novel further analysis was introduced to give growers an idea of risk. This indicated that leaf-specific weed control could offer UK leek growers an 82-86% probability of making a higher profit, and a 60% probability that that increase in profit would exceed £10000 per hectare per crop.

Action Points

The research and prototype platform produced in this project (Figure 1) should encourage growers towards a paradigm shift in their thinking about weed control. It is a win-win situation where growers could increase profits while benefiting not only environmental benefits through lower herbicide use but also potentially improving consumers' perceptions of food quality since no direct herbicide applications would be made to the crop.

It is, however, necessary to ensure expectations are realistic. The project's prototype is not a commercial product and, as of now, a considerable investment of time and money would be needed to bring the prototype platform demonstrated to market.

The project team would be interested in hearing from growers who would consider purchasing such a system and to indicate whether they would prefer a completely autonomous platform (robot) or a tractor mounted application module.

SCIENCE SECTION

Introduction

Herbicides account for 40% of the total amount of pesticides applied by vegetable growers compared to 31% and 24% for fungicides and insecticides, respectively (Garthwaite *et al.*, 2017). Weed control, apart from being important to maintain vegetable yield and quality, is becoming increasingly challenging because of UK and EU pesticide reviews and also because pesticide manufacturers are hesitant to seek to register herbicides for a market which is relatively small (Hillocks, 2012). Additionally, vegetable growers rely on a limited and old range of herbicides (first released in 60's and 70's) which require a lot of funding and effort in order to keep them in the market (Fennimore *et al.*, 2014). Legislation like the Regulation EC no. 1107/2009, the EU Water Framework Directive and the Sustainable Use of Pesticides (SUP) Directive along with UK's National Action Plan for SUP have resulted in actual losses of approval for some herbicide actives and have decreased the likelihood of new compounds gaining approval (Baker & Knight, 2017). In order to compensate for the lack of available herbicides, there has been an increase in physical and mechanical weed control methods which are often more expensive than spraying herbicides (Garthwaite *et al.*, 2017). Moreover, the environmental impact of non-chemical methods such as cultivation on soil and the soil fauna is not trivial. The option of hand weeding also exists as an expensive backstop, but may be precluded if manpower is not available for such an arduous task. These pressures act as a driver towards a paradigm shift in approaches to weed control, which will balance the need to meet demand for sustainable vegetable production while maintaining and increasing productivity (Fennimore & Cutulle, 2019). Plant- and leaf-specific weed control using herbicide droplets applied from robotic weeders offer such a paradigm shift: no herbicide to the crop, none directly to the soil, microdoses applied only to the weeds. Such a system presupposes automation.

Prerequisites for automated real-time weed control include a guidance system for navigation and to avoid crop damage, weed detection, a micro-sprayer and software to control the machine, target the weeds and an algorithm to determine whether any treatment is needed (Slaughter *et al.*, 2008b).

Detecting single weed leaves and treating them using micro-rates of foliar-applied, translocated herbicides is the ultimate in precision weed management. A study on four arable crops measured losses of pesticides up to 99% (sugar beet) to the soil surface when they are applied using broadcast spraying methods (Jensen & Spliid, 2003). Assuming that a weed seedling covers a soil surface of 1 cm^2 with numbers ranging between 100 and 400 weeds m^{-2}, this corresponds to 1%-4% weed ground cover. To put it more simply, if a foliaracting herbicide is applied by broadcast spraying, 96% will be wasted, being applied to either the soil surface or the crop. Blackmore (2013, 2014) predicted that with the use of a microdot system which sprays chemicals only on the leaf-area of the weed, should result in 99% reduction in herbicide inputs.

Nieuwenhuizen *et al*. (2010) developed a tractor-pulled microsprayer system for volunteer potato control in sugar beet fields. The system emitted single droplets (20 ± 5 µl) using 5% glyphosate solution (Roundup Max, 450 g L^{-1}). It achieved 83% volunteer potato control while spraying approximately 1% of the sugar beet plants. Crop damage was attributed to spray drift or run-off rather than inaccurate targeting. A real-time micro-sprayer, which used an inkjet printer head as a spray system was developed by Midtiby *et al*. (2011) and was tested under indoor conditions. Droplets of 0.2 µl which contained 1 µg of glyphosate were used to control two weed species at speeds of 0.5 m s^{-1}. Although the system controlled 94% of the relatively large oilseed rape plants, it only managed to control 37% of the smaller *Tripleurospermum inodorum* L. (scentless mayweed).

Miller *et al.*'s (2011) automated spot herbicide spraying system for volunteer potatoes in horticultural row crops has been commercialised. It includes a novel image analysis system to detect crop rows and weeds and specialised nozzles to spray individual weeds. Ninety to 95% weed control was achieved using glyphosate in crops of carrot, parsnip and onion. The crop damage which occurred was considered "commercially acceptable". Miller *et al.* (2013) tested the same system in leeks, achieving 95% control of volunteer potatoes, wild mint and mugwort and, importantly, they reported that no glyphosate could be detected in leek plants next to treated weeds treated.

Closer to the approach adopted here, Christensen *et al*. (2009) mentioned the use of Drop-on-Demand (DoD) technologies which apply low volume rates (approximately 1 µL) of glyphosate as a single droplet to weed leaves and have the potential of achieving herbicide savings higher than 95% when compared with broadcast application methods. The robotic DoD system described by Utstumo *et al.* (2018) demonstrated intra-row weed control in carrots using 2.1 µl droplets of glyphosate each containing 5.3 µg of glyphosate. Their results are very relevant to the eyeSpot project as the effective herbicide dose per plant in indoor conditions was 7.56 µg of glyphosate per seedling for *Chenopodium album*, *Poa annua*, *Stellaria media* and *Tripleurospermum inodorum*. The dose was applied to each seedling as three 1.16 µl droplets. However, the robotic applicator was treating with droplets all plants (weeds and carrots), no other herbicides than glyphosate were applied (either as droplets or overall spray) and no other vegetable crops were tested. Also, no yield data were recorded and it is envisaged that the robotic system would be used in conjunction with mechanical inter-row weed control.

Other autonomous platforms have been described including HortiBot (Sørensen *et al*., 2007), BoniRob with a precision spraying module (Ruckelshausen *et al*., 2009; Scholz *et al*., 2014), AgBot II with an automated selection of mechanical weeding and precision spraying depending on the weed species (Bawden *et al*., 2017), Ladybird for real-time precision spraying in vegetables (Underwood *et al*., 2015), (Bogue, 2016), and the solar-powered ecoRobotix platform (https://www.ecorobotix.com/en/).

Only Utstumo *et al.*'s (2018) system, appears to document the exact amounts of herbicide required for leaf-specific weed control using single droplets on to individual leaves. The closest to a classical dose-response relationship for droplets is by Mathiassen *et al*. (2016) who applied six doses of glyphosate (0.22 to 7 µg) as single droplets to four weed species. However, a recommended dose was not determined and the doses did not take account of the size of the weeds.

The foregoing studies largely concerned the development of the technology and none were compared with conventional sprays and weed control efficacy, crop yield, quality and gross margins/profitability were either unquantified or unclear. In order to validate the concept of weed control by leaf-specific herbicide application, glasshouse and field trials were carried out, using manual herbicide droplet applications. These were applied to selected weed species in the glasshouse or to the natural weed infestation in fields growing cabbages and leeks. Efficacy and economics of weed control and crop profitability were estimated.

Activities comprised four main areas:

1. **Glasshouse dose-response trials** with glufosinate-ammonium, glyphosate and 2,4-D on a range of weed species to evaluate amount of active ingredient required in single droplets for leaf-specific weed control. Dose-response relationships were analysed to quantify LD_{50} and LD_{90} values, i.e. doses which reduce weed biomass by 50 and 90%.
2. **Field experiments**. Leaf-specific weed control in field vegetables in transplanted cabbage and leek crops was evaluated with respect to weed control efficacy, reductions in herbicide use, crop yields and quality and profitability compared with conventional spraying.
3. **Automation.** Images of natural weed infestations in leeks and cabbages were captured automatically using a customised camera and custom-built computer system (supplied by Concurrent Solutions llc). A prototype applicator was tested to

optimise operational parameters (hydraulic pressure, distance of nozzle from target leaf) and to assess the impact of wind and wind direction on targeting accuracy. A prototype autonomous platform was developed by Concurrent Solutions in the USA (Figure 1).

4. **Dissemination.** A wide range of dissemination activities was undertaken including press reports, social media outputs and talks at both academic and trade events in the UK, Europe and further afield. These culminated in a field day was organised by AHDB in July 2019 to demonstrate operation of the prototype autonomous platform and to assess interest from the industry.

The scientific research carried out in the glasshouse and field had three main hypotheses:

1. Over 90% weed control efficacy will be achieved by applying droplets of a systemic herbicide leaf-specifically at the manufacturer's recommended rate for conventional spraying. This efficacy will equal or exceed that from conventional spraying.
2. Leaf-specific weed control will maintain or improve the yield, quality, economic value and profitability of cabbage and leek crops, compared to spraying conventional pre- and post-emergence herbicides.
3. Herbicide inputs per hectare will be much lower when weeds are controlled leaf-specifically and total amounts applied over the growing season will not exceed label approval rates.
4. Multiple leaf specific treatments will be required so that late-emerging seedlings are controlled and any failure of weed control is addressed during the critical weed-free period of the crop (Nieto *et al*., 1968).
5. Fewer droplet applications will be needed in cabbages than in leeks as leeks are known to be weak competitors against weeds.

Materials and methods

Glasshouse dose-response trials

Dose-response trials with droplets of the herbicides, glyphosate, glufosinate-ammonium and 2,4-D (**Table 1**) were carried out. Weed species tested in glasshouses at Reading University (Figure 2 A) from 2015 to 2018 were *Chenopodium album* L. (fat-hen), *Stellaria media* L. Vill. (common chickweed), *Matricaria recutita* L. (German chamomile), *Galium aparine* L. (cleavers), *Urtica urens* L. (small nettle), *Poa annua* L. (annual meadow grass), *Senecio vulgaris* L. (common groundsel) and *Rumex crispus* L. (curly dock) (Trials numbered 1-10, 12-13, Table 2). Trial 11 (**Table 2**) on *Amaranthus cruentus* L. (red amaranth) was in Kentucky in the USA in summer 2017 (Figure 2 B). Between 10 and 30 replications were tested per trial (**Table 2**) as randomized complete blocks for each weed species.

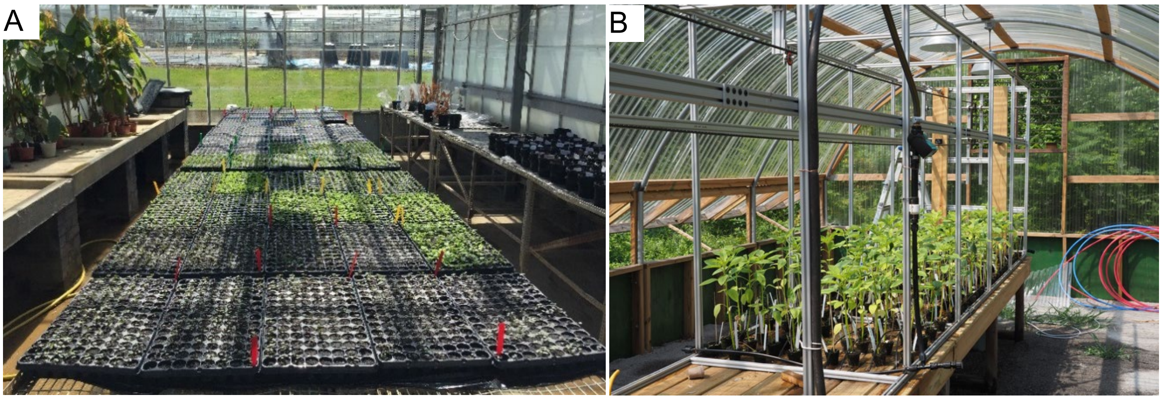


Figure 2 Glasshouses in (A) Reading and (B) Kentucky, USA. In (A), trays had 84 cells (35 mm x 35 mm x 45 mm) with one seeding per cell.

Table 1. *Details of herbicides used for the dose-response trials (adapted from Koukiasas, 2019).*

Commercial product name	Supplier	Active ingredient (ai)	Concentration, g (ai) L^{-1} (product)	Recommended dose, L (product) ha^{-1}
Roundup® Biactive GL	Monsanto (UK) Ltd.	Glyphosate	360	1.5
Harvest®	Bayer CropScience Ltd., UK	Glufosinate-ammonium	150	3
Depitox®	Nufarm (UK) Ltd.	2,4-D	500	1.4
Kyleo®	Nufarm (UK) Ltd.	2,4-D & glyphosate	160 & 240	3
Envy™ Six Max	Innvictis Crop Care, LLC™, USA	Glyphosate	540	1.18
Liberty® 280 SL	Bayer CropScience LP, USA	Glufosinate-ammonium	280	2.25

To verify the crop's susceptibility to herbicide droplets, Savoy cabbage seedlings (cv. Famosa) were tested in trial 10 (**Table 2**). Seeds of *C. album* (1974) and *R. crispus* (1988) were collected from Reading University's Farm at Sonning-on-Thames and were stored at 2-4°C. We are grateful to Herbiseed Ltd. for donating seeds of *Urtica urens*, *S. media*, *G. aparine*, *M. recutita*, *P. annua* and *S. vulgaris* and to Hammond Produce for supplying the cabbage plants*. A. cruentus seeds* were obtained from Two Willies Nursery (Lucedale, Mississippi, USA).

Table 2. *Glasshouse herbicide droplet application trials. Individual seedlings with BBCH growth stages (Feller* et al.*, 1995) and ground cover were treated with the number and volume of droplets shown. The dose of a.i. required to treat one seedling with the minimum label recommendation and the concentration of a.i. (%) in the solution in the droplet(s) to achieve that dose, are shown. Trials with the same number were concurrent. See* ***Table 1*** *for herbicide products used. Adapted from Koukiasas (2019).*

Trial (sub-trial)	Number of blocks	Plant Species	BBCH Growth Stage	Mean (sd) ground cover (cm^2) per seedling	Droplet number and volume (µL) per seedling	For minimum label recommendation (1x):	
						Solution (%)	Dose, µg
Roundup® Biactive GL (360 g L^{-1} glyphosate)							
1 (a)	30	*C. album*	12-14	1.08 (0.44)	1 x 0.648	2.5	5.83
2 (b)	10	*C. album*	14-16	7.21 (1.07)	1 x 1.082	10	38.9
3 (c)	11	*S. media*	14-16	9.03 (3.63)	1 x 1.354	10	48.8
4 (d)	17	*M. recutita*	12-14	3.13 (1.26)	1 x 0.940	5	16.9
5 (e)	15	*G. aparine*	12-14	1.56 (0.42)	1 x 0.936	2.5	8.42
6 (f)	15	*U. urens*	16-18	25.4 (3.21)	2 x 1.905	10	137.2
7 (g)	10	*P. annua*	23-24	10.54 (1.65)	1 x 1.580	10	56.9
8 (h)	10	*S. vulgaris*	12-13	2.23 (0.31)	1 x 1.340	2.5	12.1
9 (i)	25	*R. crispus*	13-14	3.28 (0.90)	1 x 0.984	5	17.7
10 (j)	29	*B. oleracea*	14-15	20.8 (3.4)	2 x 1.560	10	112.2
Envy™ Six Max (540 g L^{-1} glyphosate)							
11 (k)	10	*A. cruentus*	16-18	51.04 (13.2)	1 x 2.986	20	322.5
Harvest® (150 g L^{-1} glufosinate-ammonium)							
12 (l)	13	*C. album*	14-16	4.84 (1.56)	1 x 1.452	10	21.8
2 (m)	10	*C. album*	14-16	7.21 (1.07)	1 x 2.163	10	32.5
13 (n)	12	*U. urens*	14-16	6.25 (2.11)	1 x 1.876	10	28.1
8 (o)	10	*S. vulgaris*	12-13	2.23 (0.31)	1 x 1.340	5	10.0
7 (p)	10	*P. annua*	23-24	10.54 (1.65)	1 x 1.580	20	47.5
Liberty® 280 SL (280 g L^{-1} glufosinate-ammonium)							
11 (q)	10	*A. cruentus*	16-18	51.04 (13.2)	2 x 2.870	20	321.6
Kyleo® (160 g L-1 2,4-D and 240 g L-1 Glyphosate)							
2 (r)	10	*C. album*	14-16	7.21 (1.07)	1 x 2.163	10	86.5
8 (s)	10	*S. vulgaris*	12-13	2.23 (0.31)	1 x 1.339	5	26.8
7 (t)	10	*P. annua*	23-24	10.54 (1.65)	1 x 1.580	20	126.5
2,4-D (500 g L^{-1}) + Glufosinate-ammonium (150 g L^{-1})							
2 (u)	10	*C. album*	14-16	7.21 (1.07)	1x1.01 + 1x2.163	10+10	50.5+32.4
8 (v)	10	*S. vulgaris*	12-13	2.23 (0.31)	1x1.25 + 1x1.340	2.5+5	15.6+10
7 (w)	10	*P. annua*	23-24	10.54 (1.65)	1x1.74 + 1x1.580	20+20	174+47.5
Depitox® (500 g L^{-1} 2,4-D)							
2 (x)	10	*C. album*	14-16	7.21 (1.07)	1 x 1.010	10	50.5
8 (y)	10	*S. vulgaris*	12-13	2.23 (0.31)	1 x 1.250	2.5	15.6
7 (z)	10	*P. annua*	23-24	10.54 (1.65)	1 x 1.74	20	174

Except where noted, to apply a product's label recommendations as a single 1-2 µl droplet, a.i. concentrations required were 2.5 to 20%. Droplets were applied to the adaxial side of the youngest fully expanded leaf. In trials 6 with *U. urens* and 11 with *A. cruentus*, the larger seedlings needed two droplets to achieve the required dose (**Table 2**); these droplets were applied to the same point on the leaf. Trials 2 (u), 8 (v) and 7 (w) with the two a.i.s 2,4-D and glufosinate-ammonium, also required two droplets as a single formulation of the two a.i.s is not commercially available (**Table 2**); these droplets were applied on either side of the central vein of a single leaf.

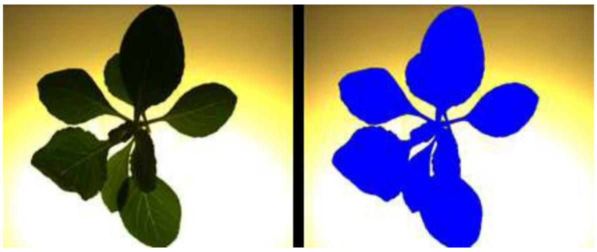


Figure 3 Images of a savoy cabbage seedling taken from above with a 90° viewing angle before (left) and after (right) image analysis to assess ground cover using WinDias software.

Herbicide doses of 1/256 to 6x the minimum label recommendation were generally applied. Recommended doses for seedlings were based on their ground cover which was estimated using photographs taken with a Nikon D90 Digital SLR Camera with an 18-105 mm VR Lens Kit, mounted on a tripod (ManFrotto Compact Action) with a 90° viewing angle. At least 10 seedlings were photographed and ground cover was estimated by the proportion of green pixels in a known area using Windias software (Figure 3, Delta T Devices Ltd.). The recommended dose for a seedling was then calculated by multiplying the ground cover in hectares by the recommended dose per hectare.

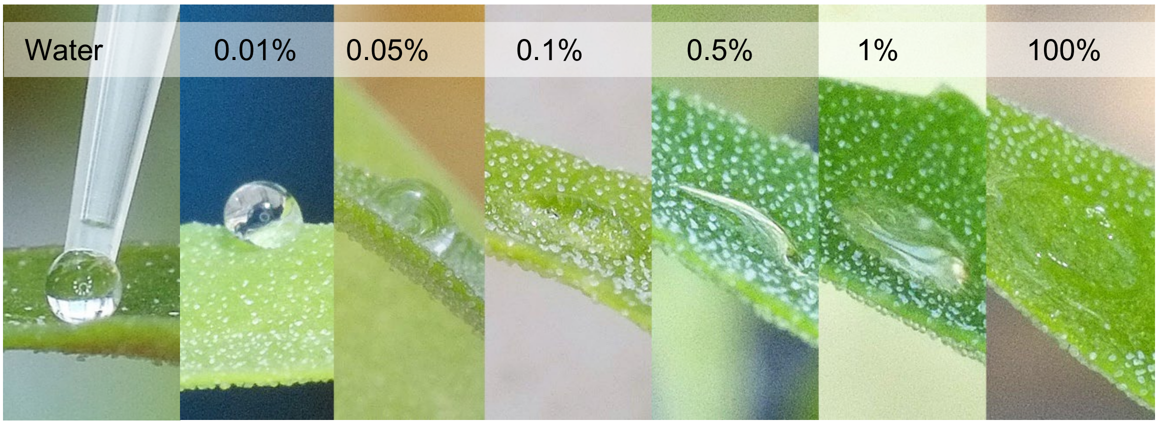


Figure 4 Droplets (1 µl) of purified water containing different concentrations of the adjuvant, AS500SL, on Chenopodium album *leaves. Water is shown on the left with the applicator tip used to apply droplets.*

Adjuvants were used in all herbicide treatments in order to achieve adequate wetting of waxy leaf surfaces. The adjuvant, AS 500 SL (Z.P.H Agromix, Niepołomice, Poland), was selected as it is particularly well-suited to glyphosate applications (Woznica *et al.*, 2015). A 1% solution was recommended by Woznica (personal communication, 2015) and provided satisfactory wetting of very waxy leaves (Figure 4). The only exception was in trial 11 (q) when a 1% solution of the adjuvant Verimax Ams Dry (Innvictis Crop Care, LLC™) was used to avoid importing pesticides to the USA.

Three control treatments were included for most dose response experiments comprising purified water, adjuvant (1%) and the undiluted herbicide product. The only exception was for 2,4-D applications to *P. annua* seedlings when the maximum label recommendation was used (3.3 L ha^{-1}) because *P. annua*, as a grass, is not expected to be susceptible to 2,4-D.

Droplets of 0.1 to 2.5 µl were applied with an ErgoOne® Single-Channel pipette (STARLAB (UK), Ltd) and droplets from 2.5-10 µl with the Micropipette Single Channel (Scilogex llc).

Fresh and dry biomass data were assessed 20 d after treatment and damage was scored using the European Weed Research Council (EWRC) scoring system (Ciba-Geigy, 1975). Dry weights were weighed to the nearest 0.0001 g after oven-drying for 48h at 80°C.

Field experiments

Transplanted cabbage and leek crops were grown at Reading University's Sonning Farm to evaluate the efficacy of leaf-specific weed control in the field. Weed control efficacy, reductions in herbicide use, crop yields and quality and the economics of weed control by different methods were all evaluated.

The profitability of leaf-specific weed control in cabbages and leeks was compared with conventional spraying. In order to help growers to have more confidence in the economic evaluation, the probability that leaf-specific weed control would be more profitable was calculated.

Field experiments were carried out at Sonning Farm, near Reading with cabbages (*Brassica oleracea* var. *capitata*) in summers 2016 and 2017, and with leeks (*Allium porrum* L.) in summers 2017 and 2018 (Figure 5 (A), **Table 3**). A leek crop planted in summer 2016 was irretrievably damaged by pests leading to the need for the 2018 experiment to achieve two years' data. Soil textures in the fields used in 2016 and 2017/2018 were classified as "sand" (91.5% sand, 5.7% clay, 2.9% silt plus 0.9% stone) and "loamy sand" in 2017 and 2018 (87.1% sand, 6.4% clay, 5.3% silt plus 12.9% stone), respectively.

In 2016, savoy cabbage seeds, were kindly provided by Elsoms Seeds Ltd (Lincolnshire, UK). They were sown in a glasshouse at Reading on Seed & Modular compost (Clover Peat, Dungannon, N. Ireland). After six weeks, the seedlings with 3-4 leaves were transplanted to the field. In 2017, white cabbage and leek seedlings were obtained from Westhorpe Plants Ltd (Boston, UK) and again were transplanted at the 3-4 leaf stage. In 2018, leek seedlings were purchased from Farringtons Ltd. (Preston, UK) and transplanted at the 2-3 leaf stage.

Fertiliser (sulfur (50 kg SO_3 ha^{-1}) and nitrogen (100 kg N ha^{-1})) was applied immediately after transplanting. Individual plants were watered twice daily for 30 min, using an automated drip irrigation system.

Field experiments were randomized complete block designs (**Table 3**). In addition to six to eight droplet or conventionally-sprayed herbicide treatments (Table 4), weedy and weed-free (hand-weeded) control plots were included in each block. Leaf-specific droplet treatments utilised Roundup® Biactive GL and Harvest® (**Table 1**) with the adjuvant AS 500 SL (1%) as for glasshouse experiments. Conventionally sprayed control plots included pendimethalin (Stomp Aqua®, 455 g a.i. L^{-1}, CS, BASF plc), applied as a pre-emergence spray and metazachlor (Sultan® 50 SC, 500 g a.i. L^{-1}, SC, Adama Agricultural Solutions UK Ltd) and bromoxynil (Buctril®, 225 g a.i. L^{-1}, EC, Bayer CropScience Ltd) as post-emergence sprays for cabbages and leeks, respectively (**Table 4**, **Table 5**). An inter-row spray of glyphosate was tested in 2016 and a 38 cm shield was used to protect the crop. All conventional sprays used a knapsack sprayer (CP 15 Electric, Cooper-Pegler, Villefranche-sur-Saone, France), with a deflector nozzle (green colour 372021, Cooper-Pegler, Villefranche-sur-Saone, France). The sprayer was calibrated to deliver 1.31 L min^{-1} using a spray volume of 200 L ha^{-1}.

Droplets (volume 2 µl) were applied with an ErgoOne® Single-Channel pipette (volume range: 0.1 to 2.5 µl; Starlab Ltd, Milton Keynes, UK; **Figure 5**(B)). For a plant-specific treatment when a single herbicide droplet was applied to each weed, droplets contained either 36 µg of glyphosate or 60 µg of glufosinate-ammonium. The latter was applied for the 2017 trial only (**Table 4**, **Table 5**). To avoid accidental crop damage, only weeds growing at least 1 cm from the crop were treated.

Multiple droplet (leaf-specific) applications were carried out according to the size of the weed or the size of individual leaves for both glyphosate and glufosinate ammonium. In 2016, droplets containing 9 µg of glyphosate were applied to a single leaf if the leaf area of the seedling was less than 1 cm^2 and, if higher, two droplets were applied to different leaves (Drop x3 gly (adj)) ((**Table 4**). However, for 2017 and 2018 trials, 9 µg of glyphosate were applied to every visible leaf with an area ≥ 1 cm^2. When dose of glufosinate-ammonium was adjusted, 7.5 µg were applied to leaves with an area ≥ 1 cm^2 ((**Table 4**, **Table 5**)).

For droplets containing 36 μg and 9 μg glyphosate, 5% and 1.25% solutions of Roundup® Biactive GL were prepared, respectively. Similarly, for glufosinate-ammonium, 60 μg and 7.5 μg droplets, were applied as 20% and 2.5% solutions of Harvest®, respectively. All droplet treatments contained 1% of the adjuvant AS 500 SL. Doses applied are based on ED_{50} and ED_{90} values which were needed to reduce the fresh weights of the *C. album* seedlings in dose-response studies in the glasshouse. The numbers of droplets applied per plot at each application time were counted, from which the application rate per hectare was calculated. Due to earlier canopy closure in the 2017 cabbage trial, the third droplet application took place six weeks after planting instead of seven as in 2016.

Crop and weed harvest

Crops and weeds were harvested from the central area of the plots (**Table 3**) where droplets had been applied. For weeds, dry biomass was measured after oven-drying for 48h at 80 °C. For the crops, the above ground fresh biomass was weighed, after which the cabbages and leeks were trimmed as for commercial sale and re-weighed. Cabbages were harvested once when the majority had reached maturity (first cabbage head splitting) and ten to twelve outer leaves were removed leaving the trimmed head. Commercially, the marketable yield of savoy and white cabbage is based on number of trimmed heads weighing more than 500 g and 1000 g respectively (A. Blair, harvesting manager TH Clements, pers. comm., 5 March 2018) although some supermarkets will sell savoy cabbages weighing 400 g per head (e.g. Aldi, UK). Because the savoy heads were trimmed very heavily and all plots and plants had to be harvested at the same time to make meaningful comparisons, the cut-off weight was lowered to 300 g per head.

Leeks were trimmed to a length of 34 cm and the stalk diameter was measured 10 cm from the base using digital callipers. The trimmed weight of leeks was separated into three categories (stalk diameter <25 mm, 25-35 mm and >35 mm). Leeks measuring <25 mm are likely to be sold for processing, whereas for the 25-35 mm and >35 mm categories are “class 1” produce and would be sold as pre-packed and loose produce, respectively (T. Casey chairman of the Leek Growers Association, pers comm., 22 August 2018).

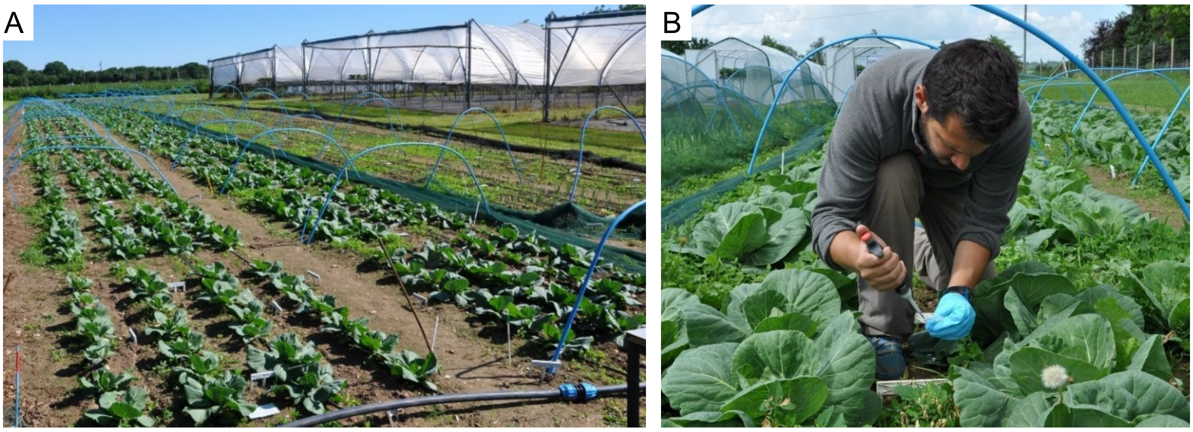


Figure 5. (A) Cabbage and leek experiments at Sonning Farm in 2017. The pipe in the foreground fed the irrigation system. (B) Manual application of a herbicide droplet with a micro-pipette in the field five weeks after transplanting the cabbage crop in 2017. The blue hoops supported netting required to protect the crop from birds.

***Table 3**. Field information and activities for the trials with cabbages (2016 & 2017) and leeks (2017 & 2018). See **Table 4** and **Table 5** for herbicide treatments.*

Activity	2016 cabbage	2017 cabbage	2017 leek	2018 leek
Field geo-reference	51°28'55"N, 0°53'51"W		51°28'24"N, 0°54'07"W	
Previous crop	Grass	Wheat	Wheat	Cabbage
Crop variety	Savoy, Famosa F1	White, Surprise F1	Krypton F1	Duraton F1
Crop planted	3 June	27 April	27 April	19 April
Crop harvested	3 October	24 July	21 August	09 August
Blocks (plots per block)	4 (8)	4 (10)	3 (9)	3 (10)
Plants planted (harvested) per plot, number	28 (8)	28 (6)	32 (8)	32 (10)
Inter-/intra-row spacing, m	0.5 / 0.3	0.5 / 0.3	0.4 / 0.2	0.4 / 0.2
Plot (Harvest & Droplet treated) areas, m^2	5.25 (1.2)	5.25 (0.9)	0.64	0.80
Insecticide	10 August: 1 kg ha^{-1} DiPel® DF	13 June: 1 kg ha^{-1} DiPel® DF	13 June: 1 kg ha^{-1} DiPel® DF	8 & 20 June: 0.8 L ha^{1} Conserve®
Fungicide	None	7 July: 1 L ha^{-1} Amistar®	None	None
Weed species (at harvest)	CAPBP, CHEAL, MATRE, SENVU, SPRAR.	ACHMI, CAPBP, CHEAL, GERMO, MATRE, POAAN, POLPE, POLAR, SENVU, TAROF, TRFDU, TRZAX.		ACHMI, CAPBP, CHEAL, MATRE, POAAN, SENVU, SOLNI, TAROF.

ACHMI: *Achillea millefolium*, CAPBP: *Capsella bursa-pastoris*, CHEAL: *Chenopodium album*, GERMO: *Geranium molle*, MATRE: *Matricaria recutita*, POAAN: *Poa annua*, POLAR: *Polygonum arenastrum*, POLPE: *Polygonum persicaria*, SENVU: *Senecio vulgaris*, SOLNI: *Solanum nigrum*, SPRAR: *Spergula arvensis*, TAROF: *Taraxacum officinale*, TRFDU: *Trifolium dubium*, TRZAX: *Triticum aestivum.*

Table 4. Herbicide treatments, doses and application times for cabbage experiments in 2016 and 2017. Droplet (Drop) treatments included adjuvant (1% AS 500 SL). Times: weeks after planting unless shown otherwise. After Koukiasas (2019).

	Treatments	Dose at each application time	Application times
2016	Drop x1 gly	36 µg of glyphosate/seedling	3
	Drop x3 gly	36 µg of glyphosate/seedling	3, 5 and 7
	Drop x3 gly (adj)	9 or 18 µg of glyphosate/seedling	3, 5 and 7
	Inter-row spray	1.5 L ha^{-1} Roundup® Biactive GL	3
	Inter-row spray + Drop x1 gly	1.5 L ha^{-1} Roundup® Biactive GL + 36 µg of glyphosate/seedling	3 (Inter-row) + 5 (Drop x 1 gly)
	Pre-emergence	2.9 L ha^{-1} Stomp Aqua®	7 d pre-planting
2017	Drop x1 gly	36 µg of glyphosate/seedling	3
	Drop x2 gly	36 µg of glyphosate/seedling	3 and 5
	Drop x3 gly	36 µg of glyphosate/seedling	3, 5 and 6
	Drop x3 gly (adj)	9 µg of glyphosate/leaf	3, 5 and 6
	Drop x3 glu-amm	60 µg of glufosinate -ammonium/seedling	3, 5 and 6
	Drop x3 glu-amm (adj)	7.5 µg of glufosinate-ammonium/leaf	3, 5 and 6
	Post-emergence	1.5 L ha^{-1} Sultan® 50 SC	4
	Pre-emergence	2.9 L ha^{-1}, Stomp Aqua®	7 d pre-planting

Table 5. Herbicide treatments, doses and application times for leek experiments in 2017 and 2018. Droplet (Drop) treatments included adjuvant (1% AS 500 SL). Times: weeks after planting unless shown otherwise. After Koukiasas (2019).

	Treatments	Dose at each application time	Application times
2017	Drop x5 gly	36 µg of glyphosate/seedling	2, 6, 8, 10 and 12
	Drop x10 gly	36 µg of glyphosate/seedling	2, 4, 5, 6, 7, 8, 9, 10, 11 and 12
	Drop x10 gly (adj)	9 µg of glyphosate/leaf	2, 4, 5, 6, 7, 8, 9, 10, 11 and 12
	Drop x10 glu-amm	60 µg of glufosinate -ammonium/seedling	2, 4, 5, 6, 7, 8, 9, 10, 11 and 12
	Drop x10 glu-amm (adj)	7.5 µg of glufosinate-ammonium/leaf	2, 4, 5, 6, 7, 8, 9, 10, 11 and 12
	Post-emergence	1.5 L ha^{-1}, Buctril®	4 and 7
	Pre-emergence	2.9 L ha^{-1}, Stomp Aqua®	5 d pre-planting
2018	Drop x5 gly	36 µg of glyphosate/seedling	3, 5, 7, 9 and 11
	Drop x5 gly (adj)	9 µg of glyphosate/leaf	3, 5, 7, 9 and 11
	Drop x10 gly	36 µg of glyphosate/seedling	3, 4, 5, 6, 7, 8, 9, 10, 11 and 12
	Drop x10 gly (adj)	9 µg of glyphosate/leaf	3, 4, 5, 6, 7, 8, 9, 10, 11 and 12
	Drop x10 glu-amm (adj)	7.5 µg of glufosinate-ammonium/leaf	3, 4, 5, 6, 7, 8, 9, 10, 11 and 12
	Post-emergence	1.5 L ha^{-1}, Buctril®	4
	Pre-emergence	2.9 L ha^{-1}, Stomp Aqua®	2 d pre-planting
	Pre + Post-emergence	2.9 L ha^{-1} Stomp Aqua® + 1.5 L ha^{-1} Buctril®	2 d pre-planting (Pre-emergence) + 4 weeks (Post-emergence)

Economic analysis

The initial economic analysis was designed to predict the profitability of leaf-specific, droplet weed control compared with conventional spraying or hand-weeding. Gross margins, which for this analysis only takes into account the costs of weed control, for droplet treatments (GM_d), conventional spraying (GM_s) and hand-weeding (GM_h) were calculated as follows:

$GM_d = V_d - H_d - M_d$
$GM_s = V_s - H_s - A_s$ and
$GM_h = V_h - L_h$

where V is the crop value (£ ha^{-1}), H is the herbicide cost (£ ha^{-1}), M_d is the annual automated droplet applicator machine cost (£ ha^{-1} yr^{-1}), A_s is the cost of using a sprayer contractor (£ ha^{-1}) and L_h is the cost of labour for hand-weeding (£ ha^{-1}). Somewhat unrealistically, the yield and the economic value of the hand-weeded (weed-free control) plots was assumed to equate with what might be achieved with commercial hand-weeding and various assumptions are made (**Table 6**). A single hand-weeding has been assumed for cabbages as the crop does not have a critical period for weed control (Weaver, 1984). By contrast three hand-weedings were assumed for leeks because the crop is only weakly competitive against weeds and the critical period of weed control is from one to 12 weeks after planting (Melander & Rasmussen, 2001; Tursun *et al.*, 2007). The costs of herbicides are based on personal communications (**Table 7**) with a contractor cost for spraying of £12.50 ha^{-1} (Redman, 2017). Other variable costs were not taken into account and have been assumed to be similar for all treatments. While higher yielding crop would cost more to harvest, it has been assumed that the cost per kg harvested was constant.

Assumptions regarding leaf-specific weed control and the automated platform were:

i. The platform treats 4 ha in an eight-hour day,
ii. Based on the field experiments and the critical periods for weed control cabbages would need to be treated three times at 14-day intervals and leeks either five times at 14-day intervals or ten times at 7-day intervals.

A sensitivity analysis of (i) was carried out to allow for weather and machine downtime, assuming the platform could operate from 1 - 7 days per week, treating 4 - 28 ha per week.

The platform cost is not known but a reasonable working assumption is around £50,000 including £10,000 for maintenance (S. Sanford, personal communication, 13 December 2018). To allow for uncertainties, a sensitivity analysis of total platform cost was also carried out assuming costs between £25,000 and £100,000. It was further assumed that this cost would be paid in equal instalments over five years.

Table 6. *Assumptions for hand-weeding costs and crop values for cabbages in 2016 and 2017) and for leeks in 2017 and 2018). Time for each hand-weeding derived from the actual time recorded hand-weeding the experimental plots over the growing season (leeks: 75 sec m^{-2} and cabbages: 46 sec m^{-2}).*

	Leeks			Cabbages		
Manual labour cost (£ h^{-1}) *	10.16			10.16		
No. of hand-weeding	3			1		
Time per hand-weeding (h ha^{-1})	208 h ha^{-1}			126 h ha^{-1}		
Total cost of hand-weeding (£ ha^{-1})	6,340			1,280		
	Stalk diameter	2017	2018		2016 (savoy)	2017 (white)
Crop value† (£ kg^{-1})	<25mm	0.50	0.60			
	=25-35mm	1.00	1.22	£ $head^{-1}$	0.42	—
	>35mm	0.82	1.00	£ kg^{-1}	0.84	0.34

*Minimum manual labour cost (Redman, 2017); † Leeks (T. Casey. pers comm., 22 August 2018), Value for cabbages is the average wholesale market prices for 2016 and 2017 (Brigham, 2017).

Table 7. *Material costs of the chemicals (herbicides and adjuvant) used for the 2017 and 2018 trials with leeks. The material cost for spraying the label recommendation glyphosate based on from and for glufosinate-ammonium costs between 6 and 10 £ ha^{-1}. The material cost of spraying pendimethalin is £20.3 ha^{-1} and for bromoxynil is £28 ha^{-1}.*

Chemicals	Cost (£ L^{-1})	Product cost to spray conventionally at rate recommended on the label	Based on personal communication in November 2018 from:
Roundup® Biactive GL	5.30	£8 to 32 ha^{-1}	Richard Casebow (Manager of Crops Research Unit, Sonning Farm)
Harvest®	2.00	£6 to 10 ha^{-1}	I. Ford (Business Development Manager, BASF Agricultural Solutions)
AS 500 SL (adjuvant)	2.59	Assuming a 1% solution in 200 L water £5.18 ha^{-1}	Prof. Zenon Woznica (Poznan University of Life Sciences)
Stomp Aqua®	7.00		Phil Lilley (Crop Production Director, Hammond Produce Ltd.)
Buctril®	19.0		Dr. Gordon Anderson-Taylor (Development Manager, Bayer CropScience Ltd.)

Table 8. *Platform costs for economic analysis including sensitivity analyses for different duty cycles (8 to 56 hours per week) and platform cost (£25-100,000). Assumed that 4 ha could be treated in an eight-hour day and that the total platform cost is spread over five years.*

			Annual cost, £ ha^{-1} year^{-1}			
			Operating days (hours) per week			
Crop	**Treatments**	**Total platform cost, £**	**1 (8)**	**3.5 (28)**	**5 (40)**	**7 (56)**
Cabbages	Droplet x3 (Fortnightly)	£25,000	£250	£71	£50	£36
		£50,000	£500	£143	£100	£71
		£75,000	£750	£214	£150	£107
		£100,000	£1,000	£286	£200	£143
Leeks	Droplet x10 (Weekly)	£25,000	£1,250	£357	£250	£179
		£50,000	£2,500	£714	£500	£357
		£75,000	£3,750	£1,071	£750	£536
		£100,000	£5,000	£1,429	£1,000	£714
	Droplet x5 (Fortnightly)	£25,000	£625	£179	£125	£89
		£50,000	£1,250	£357	£250	£179
		£75,000	£1,875	£536	£375	£268
		£100,000	£2,500	£714	£500	£357

These assumptions about the platform cost and performance give rise to predicted costs ranging from £36 to £5000 per hectare per year (**Table 8**).

Monte Carlo simulations (MCS) have rarely been used in studies of the economics of weed control (Murdoch *et al.* 2001) and were used here as a tool which might help growers in deciding whether or not to adopt the new technology. MCS evaluates the ***probability*** that leaf-specific weed control would have been more profitable than conventional spraying in the field experiments carried out. Assumptions of crop values and costs were as above, it being assumed that the platform operated for 40 hours per week at a total cost of £50,000 (£10,000 per year) leading to costs of £100, £250 and £500 ha^{-1} year^{-1} for cabbages and fortnightly or weekly treatment in leeks, respectively (**Table 8**). The MC simulations were carried out with assistance from Yiorgos Gadanakis at Reading University using the open source statistical software R, version 3.6.0 (R Development Core Team, 2018). In the MCS, 10,000 estimates of the change in profit were generated based on means and standard deviations of crop values and in each case, the difference in profit between leaf-specific control and conventional spraying was calculated. These 10,000 differences were then ranked in numerical order and to assess, for example, the likelihood that there would be no increase in profit, the ranking of the observation where the difference was zero (the break-even point), would be determined. If it were, say the 8000th MCS, then that would mean an 80% chance of making a higher profit and a 20% chance of making less profit because 8,000 MCS would have a profit difference greater than or equal to the 8000th ranked value. The mean increase in profit is difference at the 50% probability value and if both treatments were equally profitable, the break-even point would be the 5000th MCS.

Statistical analysis

Glasshouse experiments

Dose-response curves (drc) were fitted to the weed biomass data by non-linear regression using a four-parameter log-logistic model (Equation 1) using *R*, version 3.2.1 (*R* Development Core Team, 2014) and the add-on package "drc" (Ritz *et al*., 2015):

$$y = c + (d - c) / [1+\exp(b\ (\log(x) - \log(ED_{50})))] \qquad 1$$

where y is biomass, c and d are the lower and upper limits of y, respectively, b is the relative slope of the curve around the ED_{50}, x is the herbicide dose and the ED_{50} is the effective dose (ED) estimated to reduce weed biomass by 50% (Streibig,1988). Curves were usually fitted to dry weight data, but in Trial 9 (i) for *R. crispus*, the dry weight data could not be modelled according to Equation (1) and so fresh weights were analysed. The ED_{90} – the dose estimated to reduce weed biomass by 90% – was also calculated using parameter values in Equation (1) (Ritz, 2010). The lack-of-fit of Equation (1) to the data was tested by comparing the residual sum of squares (RSS) after fitting Equation (1) with the RSS of analysis of variance using the modelFit() function in *R* (Ritz & Streibig, 2012).

To explore the responses further, the reduction in weed dry biomass was calculated relative to the control using Equation (2):

$$\%\ \text{Reduction of biomass} = [1 - (W_t - W_0) / (W_c - W_0)] \times 100 \qquad (2)$$

where W_t and W_c are weed dry biomasses of individual herbicide treated and water control seedlings, respectively and W_0 is the *mean* dry biomass of a sample of seedlings on the date droplets were applied. To analyse the drc for the relative reduction of growth, the parameter c in Equation (1) was omitted (i.e. c = 0). The resulting curves were plotted against the dose relative to the herbicides' label recommendations.

The effect of the adjuvants was tested by comparing the water and adjuvant controls in a one-way analysis of variance using GenStat.

Field experiments

Crop and weed biomass data, harvest index, economic value and amounts of herbicide applied were analysed with one-way analysis of variance as a randomized complete blocks design using Genstat. Weed control efficacy was based on biomass data and expressed as per cent biomass reduction relative to the weedy control as follows:

$$(\%)\ \text{Reduction of weed biomass} = (1 - W_i / W_C) \times 100 \qquad (3)$$

where, W_i is the dry weight of any weeds surviving each weed control when the crop was harvested and W_C is the dry weight of weeds in the weedy control treatment. Yield and biomass data of the crops were expressed as a per cent relative to the weed-free control. Harvest index (HI) is the ratio of the trimmed marketable yield divided by the untrimmed biomass.

Applicator trials

Linear regressions were carried out using R version 3.2.1 (R Development Core Team, 2014) and an unbalanced ANOVA was carried out using GenStat.

Results

Glasshouse dose-response trials

Adjuvants and very low doses of herbicides had little or no effect as expected for typical dose-response curves (**Figure 6, Figure 7**). After the initial "shoulder" (**Figure 7**), increase in herbicide dose resulted in a rapid increase in the visual symptoms of phytotoxicity (**Figure 6**) and herbicide efficacy (**Figure 7**). To facilitate comparisons between weeds and herbicides, expressing application rates relative to the approved label rate shows a range of relative efficacies, ED_{50} doses being as low as $^{1}/_{93}$ and $^{1}/_{89}$ of the label-approved application rates for *Senecio vulgaris* (Trial 8h) and *Chenopodium album* (Trial 2x), with glyphosate and 2,4-D, respectively (**Table 10**). Other species and products showed a range of relative efficacies, but in 20 out of the 26 dose response trials reported here, ED_{50} doses were less than half the label rate (**Table 10**). Five of the six exceptions, where the ED_{50} dose was more than half the label rate was for Trial 7 with *Poa annua* for which the ED_{50} dose rates ranged from $^{2}/_{3}$ to nearly twice ($1^{8}/_{9}$) the label rate.

The range of phytotoxicity are exemplified in this report by the efficacies of droplet treatments applied to *Chenopodium album, Poa annua* and *Senecio vulgaris* in trials 2, 7 and 8 which included all the three herbicides and two combinations. Efficacy relative to label rates varied with species and active ingredient (Trials 2, 7 and 8: **Table 2**, **Table 10**, **Figure 6**, **Figure** 7). For example for *C. album,* 2,4-D was more effective than glyphosate or the mixture – the ED_{50} dose being, as noted above, $^{1}/_{89}$ of the label rate, compared to $^{1}/_{33}$ for the mixture with 2,4-D and $^{1}/_{6}$ for glyphosate alone, the total weight of a.i. applied per seedling also being lowest for 2,4-D (Trials 2 b/r/x: **Table 10**). For *S. vulgaris*, similar comparisons showed greater efficacy for glyphosate or the mixture than for 2,4-D (Trials 8 h/s/y: **Table 10**) while for *P. annua* the mixture was best although again more a.i. was applied than for glyphosate alone (Trials 7 g/t/z: **Table 10**). Combining glufosinate-ammonium with 2,4-D did not appear to be beneficial for any of the three species (Trials 2 m/u/x; 7 p/w/z; 8 o/v/y: **Table 10**). As indicated, ED_{50} doses were much lower than the recommended dose except for *P. annua* where the lack of efficacy in Trial 7 y for 2,4-D was expected as 2,4-D is not effective against grasses (**Table 2**,**Table** 9**, Table 10**).

When herbicide droplets were applied to different sizes of *C. album* seedlings at different times (trial numbers 1, 2 and 12), approximately 1/6th of the recommended rates of glyphosate and glufosinate-ammonium reduced biomass by 50% with the ED_{90} value being

1.5-times the recommended rate (**Table 10**).

In several trials, ED_{90} estimates were inaccurate, but where they were satisfactorily estimated, the ED_{90} rates were seldom more than the label rates (**Table 10**). It is also encouraging that efficacy was not only demonstrated for glyphosate, but also for glufosinate-ammonium and 2,4-D and combined treatment with 2,4-D and the other two actives (Trials 2, 7 and 8: **Table 2**, **Table 10**, **Figure 6**, **Figure 7**).

Additional information on these dose-response studies is provided in the project's two annual reports and in the PhD thesis co-funded by AHDB as part of the project (Koukiasas, 2019).

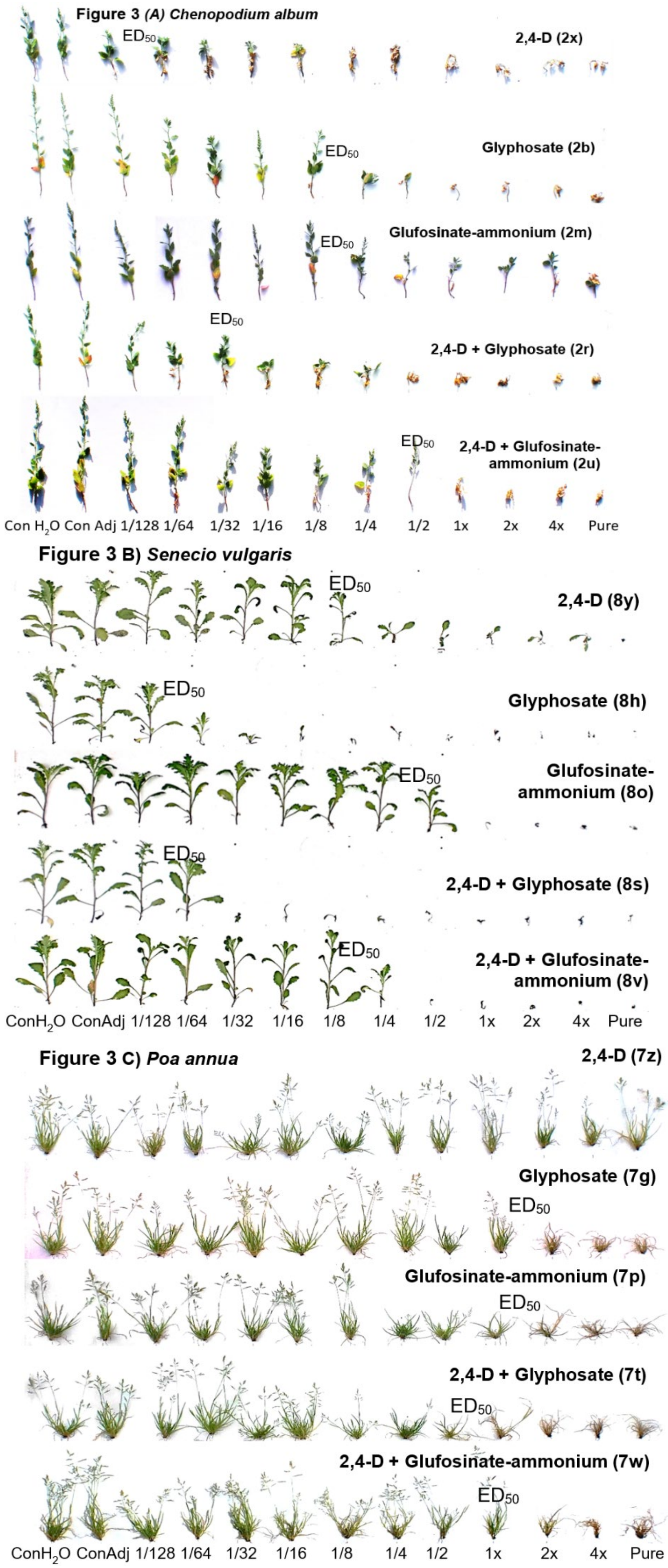


Figure 6 Seedlings of (A) Chenopodium album (Trial 2), (B) Senecio vulgaris *(Trial 8) and (C)* Poa annua *(Trial 7), three weeks after applying droplets containing doses of active ingredients relatiive to the recommended rate (**Table 2**). For the combined treatment with 2,4-D and glufosinate ammonium, two droplets were applied to the same leaf, one with each a.i. For the three controls, one droplet of deionized water (ConH$_2$O) or 1% adjuvant (ConAdj) or with undiluted herbicide (Pure) was applied. Approximate* ED$_{50}$ *doses are shown (see **Table 2** for exact estimates).*

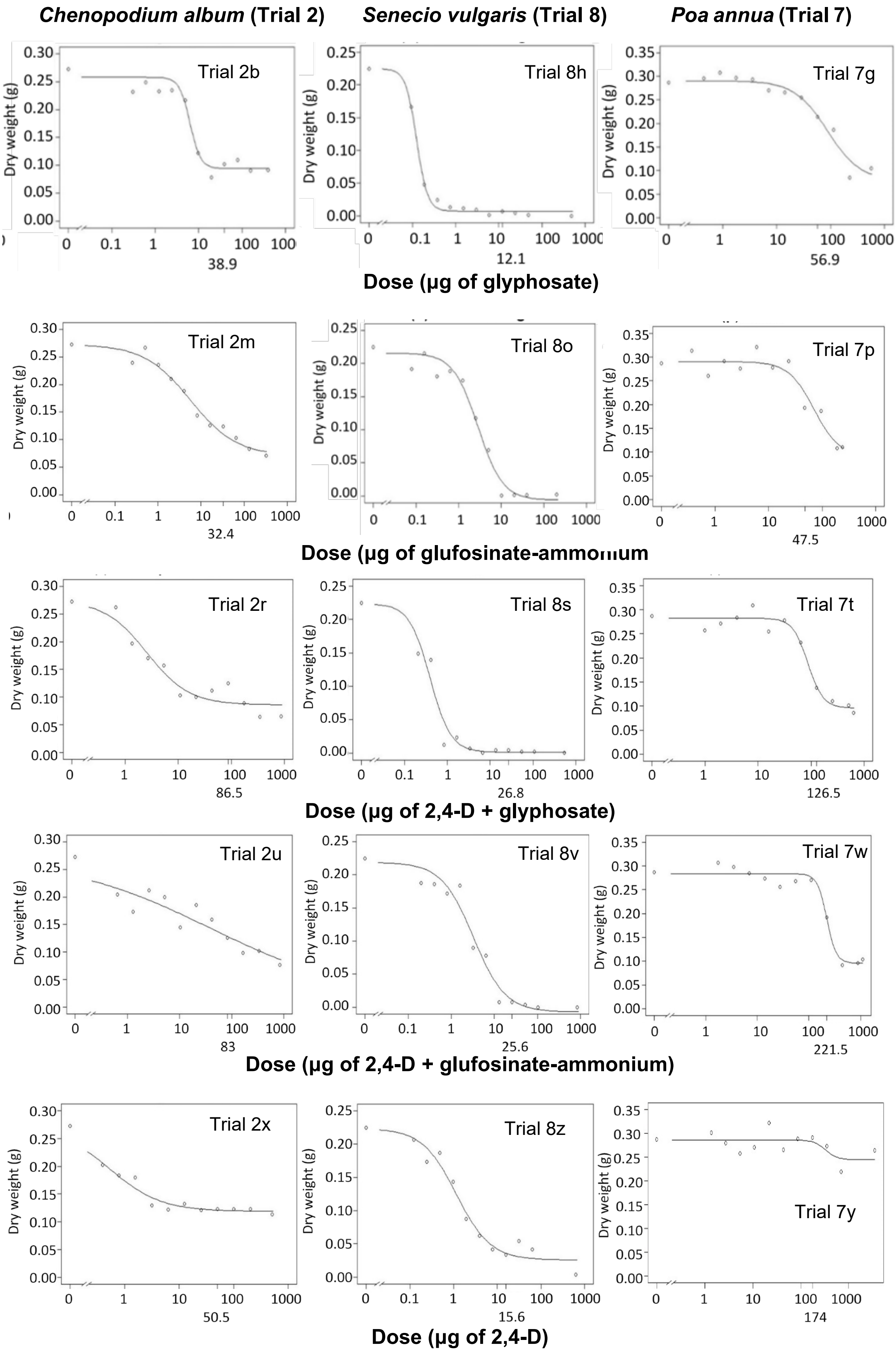


Figure 7 Dry weights of seedlings of Chenopodium album (Trial 2), Senecio vulgaris *(Trial 8) and* Poa annua *(Trial 7), three weeks after applying different doses of herbicde a.i. (**Table 2**). Curves were fitted according to Equation (1) and parameter estimates are given in **Table 9**. Recommended rates (**Table 2**) are shown below the x-axes.*

*Table 9 Parameter estimates (±SE) of fitted dose-response curves (Equation 1) for curves in Figure 7 for trials 2, 7 and 8. Doses (µg) estimated to reduce weed dry biomass by 90% (ED_{90}) 20 days after droplet application are given as are the recommended application rate per seedling at the time of droplet treatment. Application details and calculation of recommended doses applied (1x) is described in **Table 2**.*

	Parameter estimates (±SE)				Calculated values	
Trial and species	**b**	**c (g)**	**d (g)**	**ED_{50} (µg)**	**ED_{90} (µg)**	**1x rate (µg)**
Glyphosate						
(2b) *C. album*	3.54 (1.58)	0.09 (0.01)	0.26 (0.01)	6.27 (0.83)	11.7 (3.44)	38.9
(8h) *S. vulgaris*	3.39 (0.75)	0.007 (0.005)	0.22 (0.01)	0.13 (0.01)	0.24 (0.04)	12.1
(7g) *P. annua*	1.41 (0.43)	0.08 (0.03)	0.29 (0.01)	87.4 (23.2)	413 (266)	56.9
Glufosinate-ammonium						
(2m) *C. album*	0.81 (0.22)	0.07 (0.02)	0.27 (0.01)	5.66 (2.39)	84.7 (89.6)	32.4
(8o) *S. vulgaris*	1.45 (0.28)	-0.01 (0.01)	0.22 (0.01)	2.9 (0.37)	13.2 (4.3)	10.0
(7p) *P. annua*	1.82 (0.61)	0.09 (0.03)	0.29 (0.01)	70.4 (24.7)	235 (166)	47.5
2,4-D + Glyphosate						
(2r) *C. album*	1.09 (0.33	0.09 (0.01)	0.27 (0.01)	2.61 (0.68)	19.7 (14.8)	86.5
(8s) *S. vulgaris*	1.81 (0.34)	0.002 (0.005)	0.22 (0.01)	0.40 (0.04)	1.33 (0.30)	26.8
(7t) *P. annua*	3.01 (1.10)	0.09 (0.01)	0.28 (0.01)	86.7 (11.6)	180 (58.8)	126.5
2,4-D + Glufosinate-ammonium						
(2u) *C. album*	0.31 (0.07)	0.02 (0.04)	0.27 (0.01)	35 (40)	39209 (93409)	82.9
(8v) *S. vulgaris*	1.20 (0.20)	-0.01 (0.01)	0.22 (0.01)	3.26 (0.46)	20.4 (7.15)	25.7
(7w) *P. annua*	4.83 (3.89)	0.09 (0.01)	0.28 (0.01)	222 (18.7)	349 (126)	221.4
2,4-D						
(2x) *C. album*	1.05 (0.41)	0.12 (0.01)	0.27 (0.01)	0.57 (0.19)	4.60 (3.68)	50.5
(8y) *S. vulgaris*	1.12 (0.23)	0.03 (0.01)	0.22 (0.01)	1.16 (0.19)	8.21 (3.71)	15.6
(7z) *P. annua*	3.56 (3.27)	0.24 (0.02)	0.28 (0.01)	329 (167)	610 (337)	174

Table 10. *Doses of glyphosate, glufosinate-ammonium, 2,4-D, 2,4-D + glyphosate and 2,4-D + glufosinate-ammonium estimated to reduce weed biomass by 50 (ED_{50}) and 90% (ED_{90}), 20 days after droplet application. Estimates are expressed both as weight of a.i. to be applied to each seedling and this weight as a fraction of the application rate recommended on the product label.* ED_{50} *estimates calculated using Equation (1).*

Trial (sub-trial)	Plant Species	ED_{50}, µg per seedling (±SE)	ED_{90}, µg per seedling (±SE)	Recommended (1x) dose of a.i., µg	ED_{50} rate relative to 1x rate	ED_{90} rate relative to 1x rate
		Roundup® Biactive GL (360 g L^{-1} glyphosate)				
1 (a)	*C. album*	1.39 (0.56)	12.1 (8.06) *	5.83	¼	2 *
2 (b)	*C. album*	6.27 (0.83)	11.7 (3.44)	38.9	$^1/_6$	$^1/_3$
3 (c)	*S. media*	3.04 (1.10)	6.33 (7.83) *	48.8	$^1/_{16}$	$^1/_8$ *
4 (d)	*M. recutita*	2.07 (0.63)	13.6 (8.9) *	16.9	$^1/_8$	$^4/_5$ *
5 (e)	*G. aparine*	6.89 (2.15)	27.1 (20.8) *	8.42	$^5/_6$	3 *
6 (f)	*U. urens*	46.5 (15.9)	460 (389) *	137.2	$^1/_3$	3 *
7 (g)	*P. annua*	87.4 (23.2)	413 (266) *	56.9	1½	7 *
8 (h)	*S. vulgaris*	0.13 (0.01)	0.24 (0.04)	12.1	$^1/_{93}$	$^1/_{50}$
9 (i)	*R. crispus*	3.70 (3.27)	168 (340) *	17.7	$^1/_5$	9½ *
10 (j)	*B. oleracea*	35.9 (7.99)	346 (171)	112.2	$^1/_3$	3
		Envy™ Six Max (540 g L^{-1} glyphosate)				
11 (k)	*A. cruentus*	10.9 (1.10)	33.5 (7.1)	322.5	$^1/_{30}$	$^1/_9$
		Harvest® (150 g L^{-1} glufosinate-ammonium)				
12 (l)	*C. album*	4.43 (1.21)	8.91 (5.93 *)	21.8	$^1/_5$	$^2/_5$ *
2 (m)	*C. album*	5.66 (2.39)	84.7 (89.6) *	32.5	$^1/_6$	2½ *
13 (n)	*U. urens*	1.25 (0.17)	2.60 (0.78)	28.1	$^1/_{22}$	$^1/_{11}$
8 (o)	*S. vulgaris*	2.9 (0.37)	13.2 (4.3)	10.0	$^2/_7$	$1^1/_3$
7 (p)	*P. annua*	70.4 (24.7)	235 (166) *	47.5	1½	5 *
		Liberty® 280 SL (280 g L^{-1} glufosinate-ammonium)				
11 (q)	*A. cruentus*	37.5 (14.5)	1453 (1466) *	321.6	$^1/_9$	4½ *
		Kyleo® (160 g L^{-1} 2,4-D and 240 g L^{-1} Glyphosate)				
2 (r)	*C. album*	2.61 (0.68)	19.7 (14.8) *	86.5	$^1/_{33}$	$^2/_9$ *
8 (s)	*S. vulgaris*	0.40 (0.04)	1.33 (0.30)	26.8	$^1/_{67}$	$^1/_{20}$
7 (t)	*P. annua*	86.7 (11.6)	180 (58.8)	126.5	$^2/_3$	$1^3/_7$
		2,4-D (500 g L^{-1}) + Glufosinate-ammonium (150 g L^{-1})				
2 (u)	*C. album*	35 (40)	39209 (93409) *	82.9	$^3/_7$	473 *
8 (v)	*S. vulgaris*	3.26 (0.46)	20.4 (7.15)	25.7	$^1/_8$	$^4/_5$
7 (w)	*P. annua*	222 (18.7)	349 (126)	221.4	1	$1^4/_7$
		Depitox® (500 g L^{-1} 2,4-D)				
2 (x)	*C. album*	0.57 (0.19)	4.60 (3.68) *	50.5	$^1/_{89}$	$^1/_{11}$ *
8 (y)	*S. vulgaris*	1.16 (0.19)	8.21 (3.71)	15.6	$^1/_{13}$	½
7 (z)	*P. annua*	329 (167)	610 (337) *	174	$1^8/_9$	3½ *

* NB. Estimate of ED_{90} is uncertain: SE is more than half the estimated ED_{90} value.

Field experiments

Efficacy of weed control

Visual inspection of the weedy control plots showed that there was a heavy natural weed infestation in both years which was controlled well by hand-weeding and by the droplet applications and to a lesser extent by the pre-emergence sprays (**Figure 8**) especially in leeks (Figure 9).

These visual impressions were quantified by measuring the dry weights of weeds in harvested areas at the same time as harvesting the crops and estimating the efficacy of weed control using these measurements.

In cabbages, when one glyphosate droplet was applied to each weed present on three application times (Drop x3 gly) weed control efficacies were 92 and 93% in 2016 and 2017, respectively (Figure 11). Weeds were also controlled with 89 and 91% efficacies in treatments adjusted (‘adj’) according weed size in 2016 or individual leaf area in 2017, respectively. Control was much poorer if crops were treated once only (Figure 11). The pre-emergence conventional spray with pendimethalin gave variable control, achieving only 62% efficacy in 2016 but 88% in 2017, while post-emergence treatment with metazachlor achieved only 68% weed control in 2017 (Figure 11).

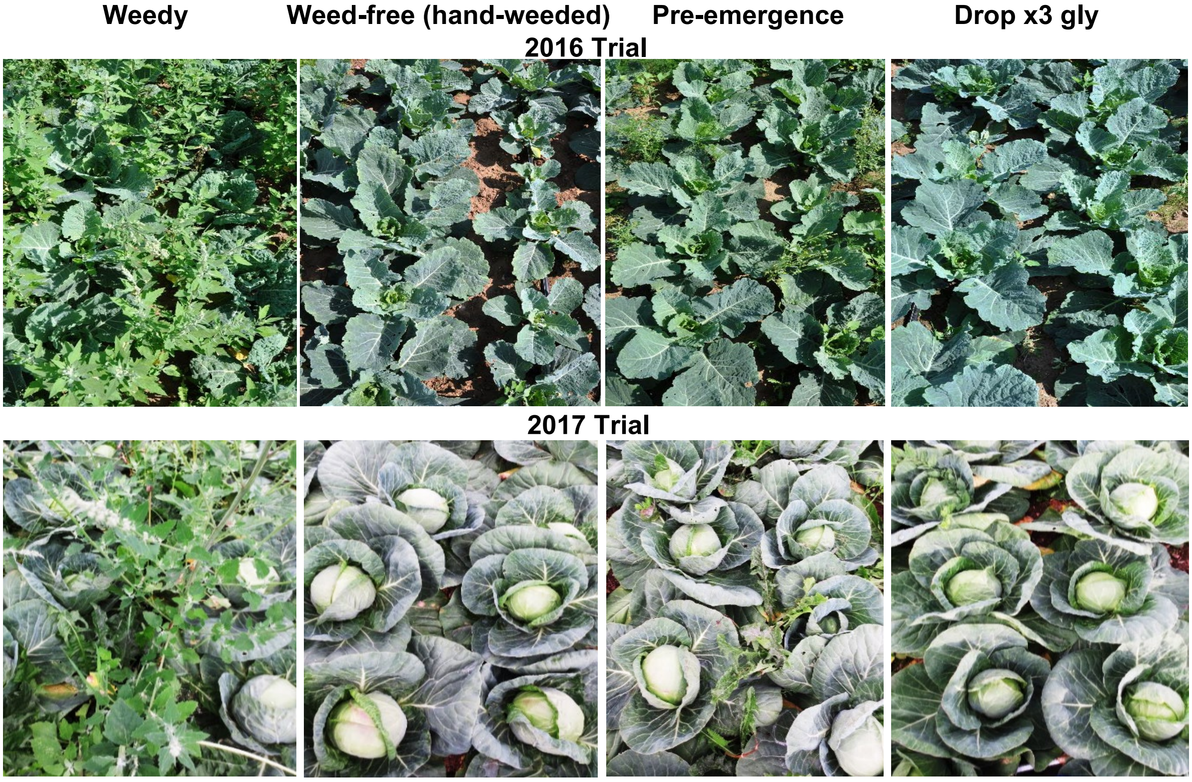


Figure 8. *Examples of plots in the 2016 and 2017 field trials with cabbages. Plots shown are the Weedy, Weed-free, Pre-emergence controls and the Drop x3 gly treatments seven and nine weeks after transplanting in 2016 and 2017, respectively. Images have been cropped to show the areas in each plot from which cabbages and weeds were harvested and to which droplets were applied in the Drop x3 gly treatment.*

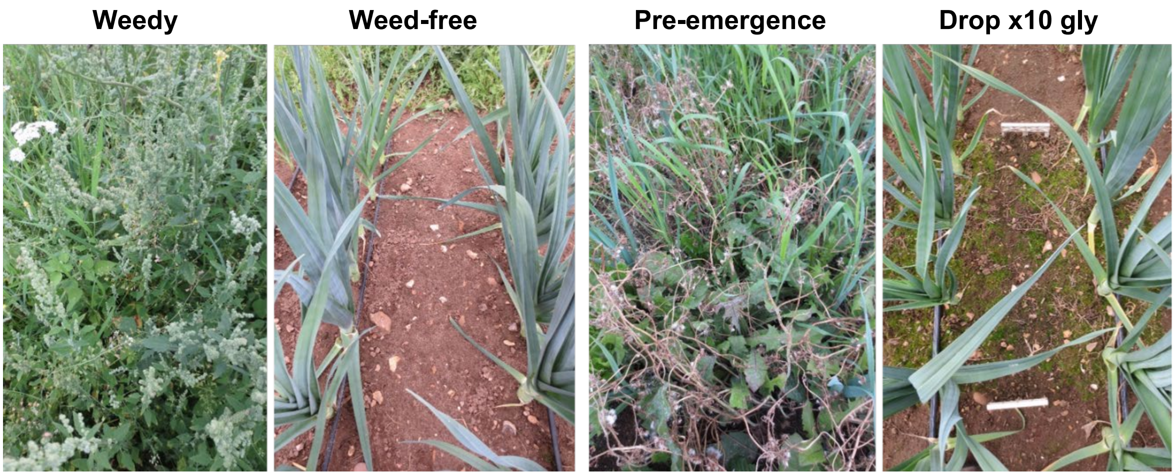


Figure 9 Examples of plots in the 2017 field trial with leeks. Plots shown are the Weedy, Weed-free, Pre-emergence controls and the Drop x10 gly treatments eleven weeks after transplanting the leeks. Images have been cropped to show the areas in each plot from which leeks and weeds were harvested and to which droplets were applied in the Drop x10 gly treatment.

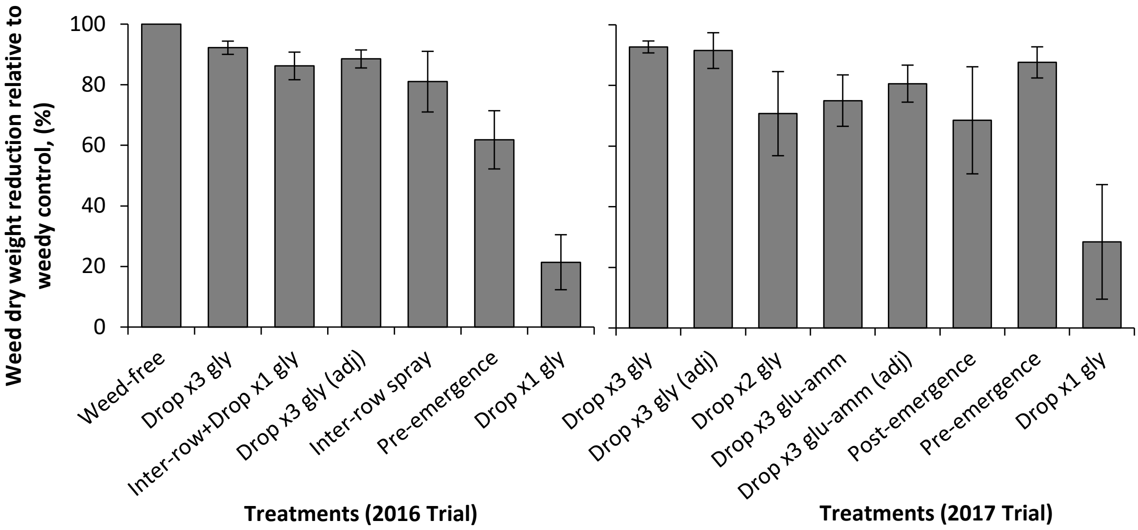


Figure 11 Weed control efficacy in cabbage field trials. Efficacy is expressed as the reduction in weed dry biomass relative to the dry biomass in the weedy control plots, i.e. 244 and 393 g m^{-2} in 2016 and 2017, respectively. Adjusted (adj) treatment: in 2016 weed seedlings received 9 or 18 µg of glyphosate if the total leaf area of a weed was less or more than 1 cm^2; in 2017, each leaf >1 cm^2 in area received 9 µg of glyphosate (gly (adj)) or 7.5 µg of glufosinate-ammonium (glu-amm (adj)). Error bars are SEMs. SEDs from analyses of variance were 11.6 (d.f.: 21) and 15.9% (d.f.: 27) in 2016 and 2017, respectively.

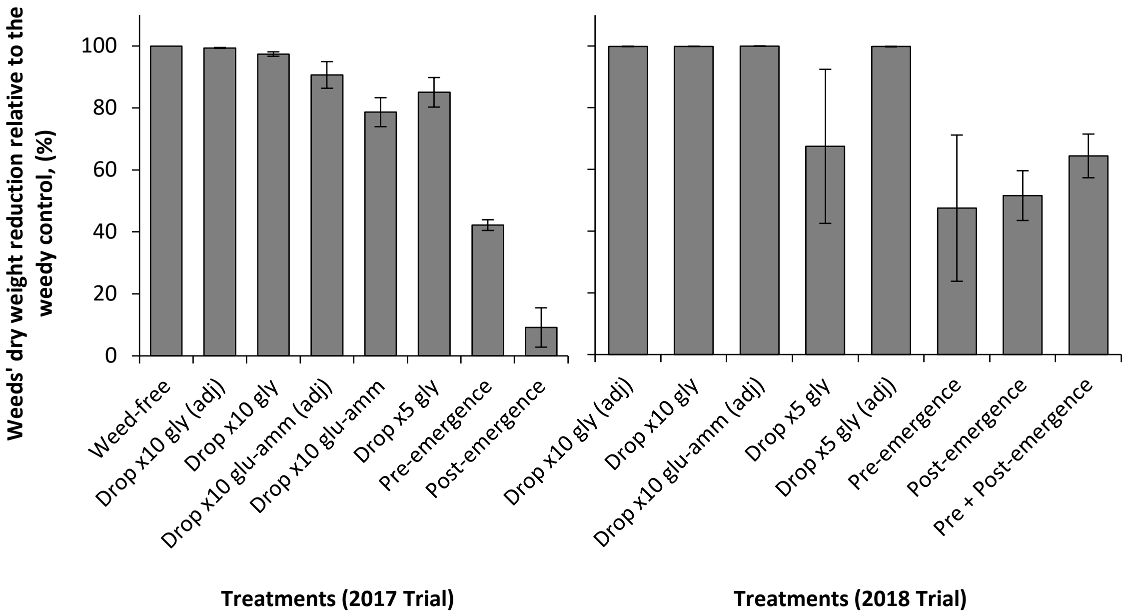


Figure 10 Weed control efficacy in leek field trials. Efficacy is expressed as the reduction in weed dry biomass relative to the dry biomass in the weedy control plots, i.e. 537 and 547 g m^{-2} in 2017 and 2018, respectively. In adjusted treatments (adj), each leaf >1 cm^2 in area received 9 µg of glyphosate or 7.5 µg of glufosinate-ammonium.). Error bars are SEMs. SEDs from analyses of variance were 11.6% (d.f.: 21) and 15.9% (d.f.: 27) in 2017 and 2018, respectively.

In leeks, the efficacy of weed control exceeded 97% when glyphosate droplets were applied ten times over 12 weeks either as one droplet per weed or to every weed leaf >1 cm^2 (Figure 10). Effective weed control was achieved with the fortnightly glyphosate droplet treatment to every weed leaf >1 cm^2 in 2018 (Figure 10). Glufosinate-ammonium was also effective as an adjusted treatment, reducing weed dry biomass by 91% and 99% in 2017 and 2018, respectively (Figure 10). Efficacy of weed control from the spray applications of pre and post-emergence herbicides was significantly lower ($P<0.001$) than the hand-weeded controls for both years (Figure 10).

Crop yields

Cabbages varied in size considerably according to weed control treatments in 2016 but not in 2017 (**Figure 12**). These visual impressions were confirmed when the trimmed and marketable yields and harvest indices were measured and analysed ($P\leq0.01$, **Figure 13**). In 2016, the weed-free control yield of savoy cabbages was 50.5 t ha^{-1} which was not significantly different from the trimmed and marketable yields from the single and multiple droplet treatments and the combined inter-row spray plus single droplet treatment (**Figure 13**). Marketable yields from the pre-emergence spray of pendimethalin and inter-row spray of glyphosate for the 2016 trial were not significantly different than the hand-weeded control (**Figure 13**). Overall 36% of the harvested savoy heads were classified unmarketable (head weight < 300 g) whereas for the 2017 trial the overall rejection rate of unmarketable white cabbages was only 11%. In 2017, trimmed and marketable yields of the white cabbages were much higher (93.5 and 88.5 t ha^{-1} in the weed-free control) but no significant differences occurred between weed control treatments for trimmed ($P=0.26$) or marketable ($P=0.24$) yields or for harvest index ($P=0.08$). However, from the producers' perspective, it is worth noticing that the lowest marketable yields occurred when droplets of glyphosate were applied on only one occasion (**Figure 13**).

Yields of trimmed leeks when approximately weekly glyphosate droplets were applied to every weed leaf (the adjusted treatment), were the highest among the plots treated with herbicides whether leaf-specifically as droplets or by conventional spraying and were not lower than the hand-weeded controls (2017: 42 t ha^{-1}; 2018: 39 t ha^{-1}; **Figure 14**). Similarly, the yield for the weekly application of the adjusted leaf-specific glufosinate-ammonium treatment was 87% of the weed-free control yield in 2018 (**Figure 14**). However, yield and weed control efficacy of the unadjusted, plant-specific glufosinate-ammonium treatment was lower than the weed-free controls and therefore it was not repeated in the 2018 trial (**Figure 14**). Yields from all the other herbicide treatments applied as droplets or overall spray were lower than the weed-free controls for both years ($P<0.05$, **Figure 14**).

__Figure 12.__ Trimmed savoy and white cabbage heads at harvest in 2016 (upper panel) and 2017 (lower panel) trials, for the treatments used each year. The head of weed-free for the 2016 trial is not a representative size of the heads harvested from this treatment.

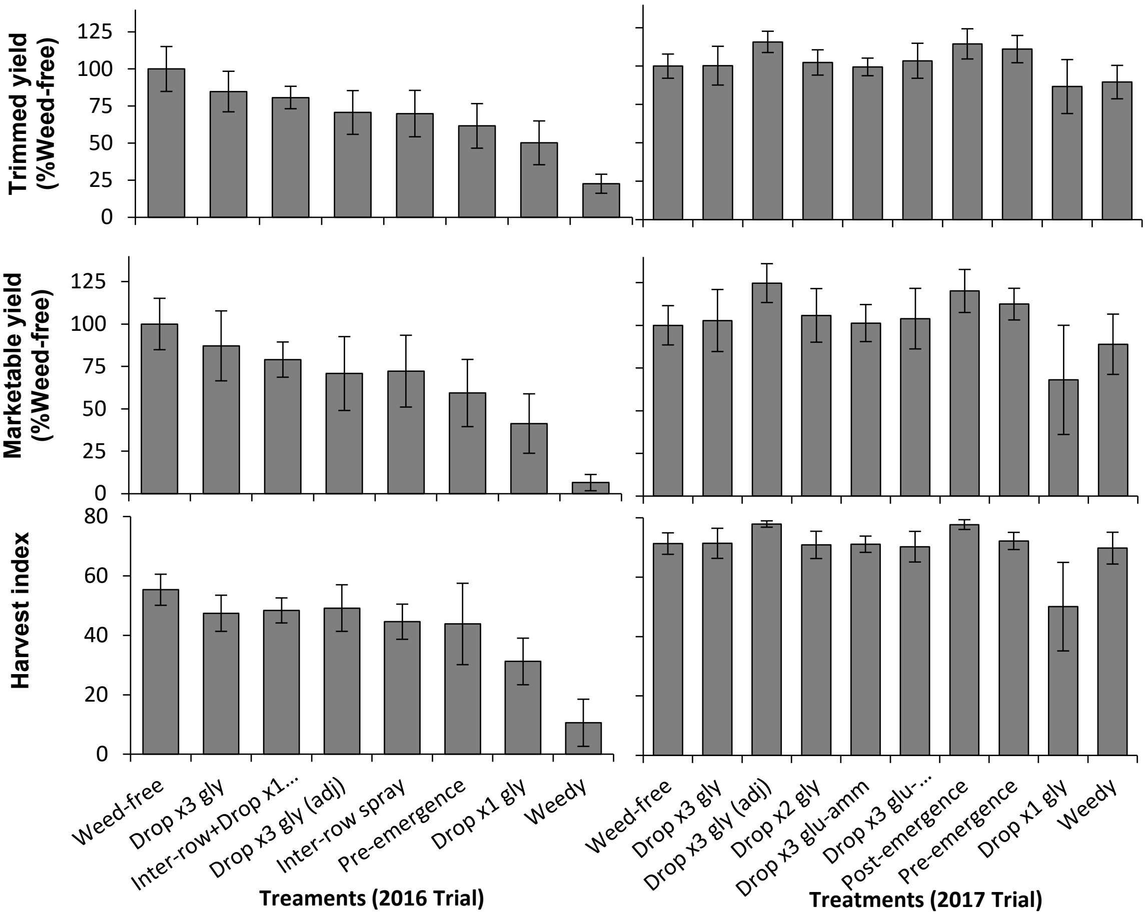


__Figure 13.__ Trimmed and marketable yields and harvest indices of cabbages under different weed control regimes. Trimmed (marketable) yields are percentages of weed-free yields (2016: 50.5 (50.5) and 2017: 93.5 (88.5) t ha^{-1}). In 2016 and 2017, respectively, SEDs (d.f.) were 16.7 % (21) and 11.8 % (27) for trimmed yields, 21.6 % (21) and 19.1 % (27) for marketable yields and 10.5 (21) and 7.7 (27) for harvest indices. Bars show means ±SEM.

Optimal treatments which achieved high levels of weed control (> 97% efficacy) and high yields produced more leeks with stalks measuring more than 25 mm (class 1 marketable yield). The mean harvest indices were 65% and 72% in 2017 and 2018, respectively and no significant differences occurred between treatments in either year (2017: P=0.9, 2018: P=0.7, **Figure 14**))

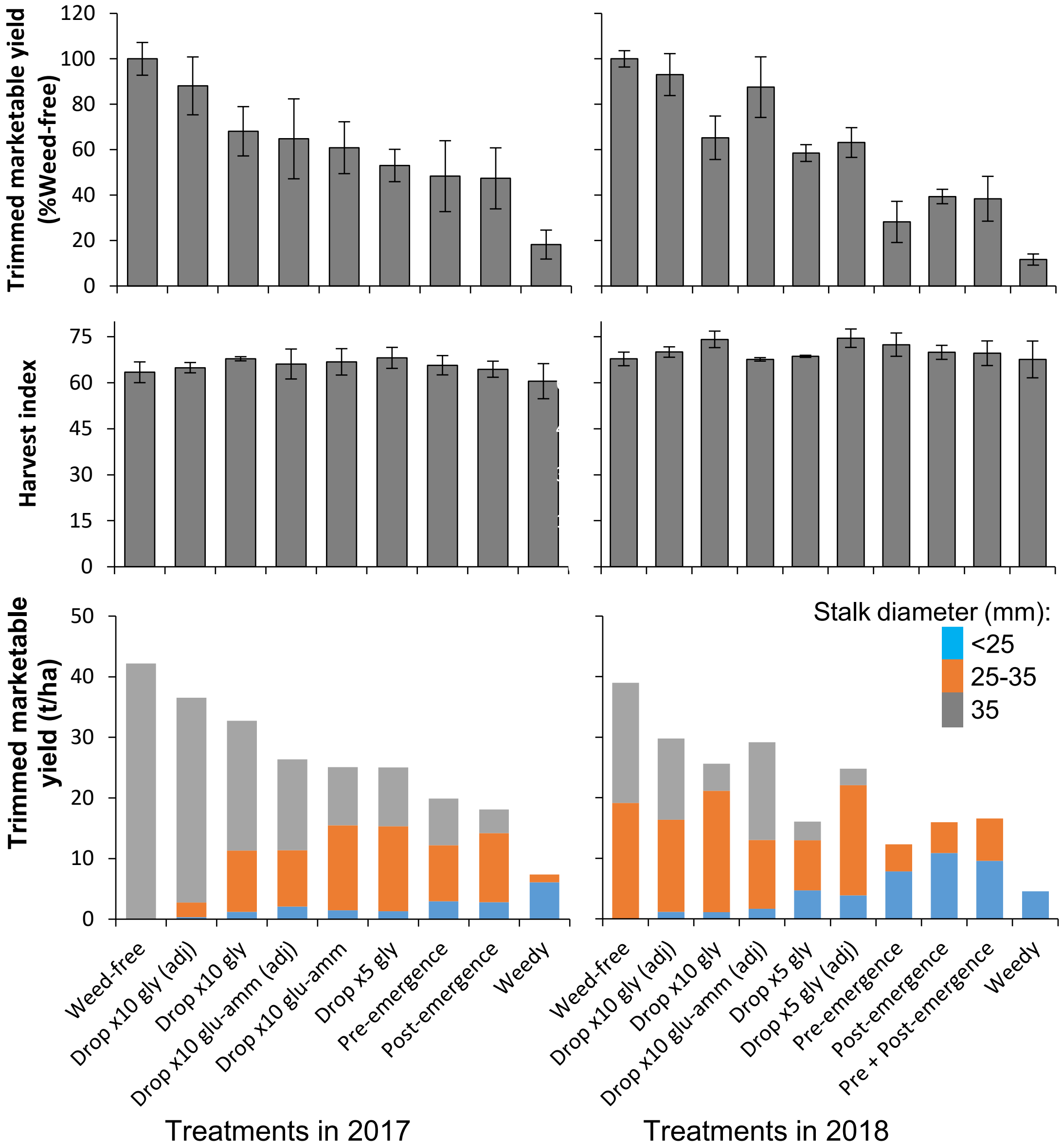


__Figure 14.__ Trimmed marketable yields and harvest indices of leeks under different weed control regimes. Yields in the top panels are expressed as percentages relative to the weed-free yields (42.2 and 39 t ha^{-1}, in 2017 and 2018, respectively). In the bottom panels, actual yields (t ha^{-1}) are classified by stalk diameter (<25mm, 25-35mm and >35 mm). Harvest index is the ratio of the trimmed marketable yield divided by the untrimmed biomass. In 2017 and 2018, respectively, SEDs (d.f.) were 11.0 % (16) and 10.5 % (18) for yields, and 4.88 (16) and 4.63 (18) for harvest indices. Bars show means ±SEM.

Herbicide use

Leaf and plant-specific weed control, greatly reduced herbicide use in cabbages. The optimal glyphosate droplet treatments (Drop x3 gly and Drop x3 gly (adj)) not only achieved high levels of weed control without significant yield penalties, but also reduced the use of herbicide actives relative to the pendimethalin spray by at least 91% and 96% for the 2016 and 2017 trials respectively (Table 3.7). When the dose of glufosinate-ammonium was adjusted for the 2017 trial it reduced herbicide use by 95% and 97% relative to the post-emergence spray of metazachlor and the pre-emergence spray respectively.

Table 11. *Mean total amounts (±SEM) of herbicide applied to cabbage plots (g a.i. ha^{-1}) for the droplet treatments and reductions relative to the pre-emergence and post-emergence spray treatments. Label recommendation for pre-emergence spray of pendimethalin is 1320 g ha^{-1}. For spraying glyphosate, the label recommendations range from 540 to 1800 g ha^{-1} and for glufosinate-ammonium range from 450 to 750 g ha^{-1} with a maximum of 1500 g ha^{-1} per year if two treatments are applied.*

Treatments	**Total amount of herbicide applied, g a.i. ha^{-1}**	**Change in relative amount of a.i. used, %, compared with**	
		Pre-emergence spray, 1320 g pendimethalin ha^{-1}	**Post-emergence inter-row spray, 540 g glyphosate ha^{-1}**
	2016		
Drop x1 gly	53.9 (7.4)	**-95.9** (0.6)	**-90.0** (1.4)
Drop x3 gly	83.3 (11.7)	**-93.7** (0.9)	-84.6 (2.2)
Drop x3 gly (adj)	119 (29.3)	**-91.0** (2.2)	-77.9 (5.4)
Inter-row spray + Drop x1 gly	562 (3.2)	-57.4 (0.2)	+ 4.1 (0.6)
SED (d.f.)	17.7 (9)	1.3 (9)	3.3 (9)
	2017		**Broadcast spray (metazachlor, 750 g ha^{-1})**
Drop x1 gly	16.4 (5.3)	**-98.8** (0.4)	**-97.8** (0.7)
Drop x2 gly	41.0 (13.4)	**-96.9** (1.1)	**-94.5** (1.8)
Drop x3 gly	55.2 (17.9)	**-95.8** (1.4)	**-92.6** (2.4)
Drop x3 gly (adj)	28.1 (9.9)	**-97.9** (0.7)	**-96.3** (1.3)
Drop x3 glu-amm	105 (42.3)	**-92.1** (3.2)	-86.1 (5.6)
Drop x3 glu-amm (adj)	40.2 (17.6)	**-97.0** (1.3)	-94.6 (2.4)
SED (d.f.)	24.2 (15)	1.8 (15)	3.2 (15)

Table 12. *Total number of droplets per square metre and amounts of herbicide applied over the growing season for the plant/leaf specific weed control of leeks in 2017 and 2018. Total amounts (g a.i. ha^{-1}) are given along with and differences relative to the pre-emergence (1319.5 g ha^{-1} pendimethalin) and post-emergence (337.5 g ha^{-1} bromoxynil) spray and the combined treatment of the two herbicides. In 2017, no combined treatment was applied and two spray applications of the post-emergence treatment were carried out (675 g ha^{-1} bromoxynil). For spraying glyphosate, the label recommendations range from 540 to 1800 g ha^{-1} and for glufosinate-ammonium range from 450 to 750 g ha^{-1} with a maximum of 1500 g ha^{-1} per year if two treatments are applied. Figures are means (±SEM).*

Treatments	Droplets applied, number m^{-2}	Herbicide applied (g a.i. ha^{-1})	Change in relative amount of a.i. used, %, compared with sprays applied: Pre-emergence	Post-emergence	Pre + Post-emergence
		2017			
Drop x5 gly	1944	700 (139)	-47.0 (10.6)	+3.69 (20.6)	N.A.
Drop x10 gly	2584	930 (34.8)	-29.5 (2.64)	+37.8 (5.16)	N.A.
Drop x10 gly (adj)	3781	340 (26.2)	**-74.2** (1.99)	-49.6 (3.9)	N.A.
Drop x10 glu-amm	,,,	2120 (140)	+60.7 (10.6)	+214 (20.7)	N.A.
Drop x10 glu-amm (adj)	8614	646(123)	-51.0 (9.29)	-4.29 (18.2)	N.A.
SED (d.f.)	…	126 (8)	9.52 (8)	18.6 (8)	N.A.
		2018			
Drop x5 gly	1590	572 (26.4)	-56.6 (2)	+69.6 (7.83)	-65.5 (1.59)
Drop x5 gly (adj)	4187	377 (24)	**-71.4** (1.82)	+11.6 (7.12)	**-77.3** (1.45)
Drop x10 gly	2242	807 (11.6)	-38.8 (8.46)	+139.1 (33.1)	-51.3 (6.74)
Drop x10 gly (adj)	3310	299 (47.4)	**-77.3** (3.56)	-11.3 (14.1)	**-81.9** (2.86)
Drop x10 glu-amm (adj)	5676	426 (18.8)	-67.7 (1.42)	+26.1 (5.56)	**-74.3** (1.13)
SED (d.f.)	…	66.4 (8)	5.03 (8)	19.7 (8)	4.01 (8)

N.A.: not applicable. … not available

For leeks, droplet applications commenced three weeks after planting in 2018 compared to two in 2017. More droplets were applied in 2017 compared to 2018 and, for adjusted treatments in a given year, more glufosinate-ammonium droplets were required compared to glyphosate (**Table 12**). Peak times for droplet applications were 6 to 8 weeks after planting. Regarding the amounts of herbicide applied, the adjusted droplet treatment of glyphosate received 340 and 299 g of glyphosate ha^{-1} at the rate of 3781 and 3310 droplets m^{-2}h in 2017 and 2018, respectively. These treatments reduced the amount of a.i. per hectare by 74% to 77% compared with the pre-emergence conventional spray and 82% lower than spraying both pre and post-emergence herbicides in 2018 (**Table 12**). The fortnightly adjusted application of glyphosate in 2018 also reduced the herbicide inputs by over 70% compared to the conventional sprays (**Table 12**). 71% and 77% relative to pre-emergence and the pre+post-emergence sprays, respectively. Weekly adjusted (leaf-specific) application of glufosinate-ammonium droplets also reduced herbicide a.i. application, by 51% and 74% in 2017 and 2018 trials, respectively, but when applied plant-specifically, as a single droplet per weed in 2017, the maximum approved label rate of 1500 g ha^{-1} was exceeded and the application of 2120 g ha^{-1}) was 61% more than the conventional pre-emergence treatment (**Table 12**). With this exception, the amounts of a.i. applied per unit land area never exceeded label recommendations for conventional spraying.

Economic analysis

In cabbages, the fortnightly plant-specific glyphosate treatment resulted in gross margins of about £32,000 ha^{-1} in both years ("Drop x3 gly": Figure 15 A, B). In 2016, only this treatment (Drop x3 gly) and the combined inter-row spray plus droplet application were not significantly lower than the weed-free (£42,441 ha^{-1}) control for the savoy cabbages. In 2017, no significant differences were observed for the economic value among the treatments applied for the 2017 trial with white cabbages (P=0.13, Appendix). The gross margin of the 'conventional' pre-emergence spray was therefore similar to the droplet treatments (P>0.05, compare "Drop x3 gly" and "pre-em" in Figure 15A, B) although there was some evidence of a trend in 2016 when the "pre-em" gross margin was only £21,877 ha^{-1}.

Leek yields were higher in 2017 than in 2018, but because of the differences in pricing between years, the crop value was higher in 2018. When droplets of glyphosate were applied weekly either plant specifically as a single droplet or leaf-specifically as multiple droplets per weed, The value of the crop was not significantly lower than the weed-free treatment (2017: £34,602 ha^{-1} and 2018: £43,187 ha^{-1}) (Figure 14). Weekly leaf-specific (x10, adj) applications of glyphosate in 2017 and 2018 and of glufosinate-ammonium in 2018, also achieved gross margins of £29-32,000 ha^{-1}, and these were much higher than the £10-17,000 ha^{-1} calculated for 'conventional' pre-emergence spraying (P<0.05, Figure 15C, D).

Using the results from Figure 15 C and D for leeks in 2017 and 2018, Monte Carlo simulations were carried out to provide what is believed might be a more interesting and relevant way of making comparisons of the profitability of different weed control options. Rather than a conventional statistical comparison, the means and standard deviations of

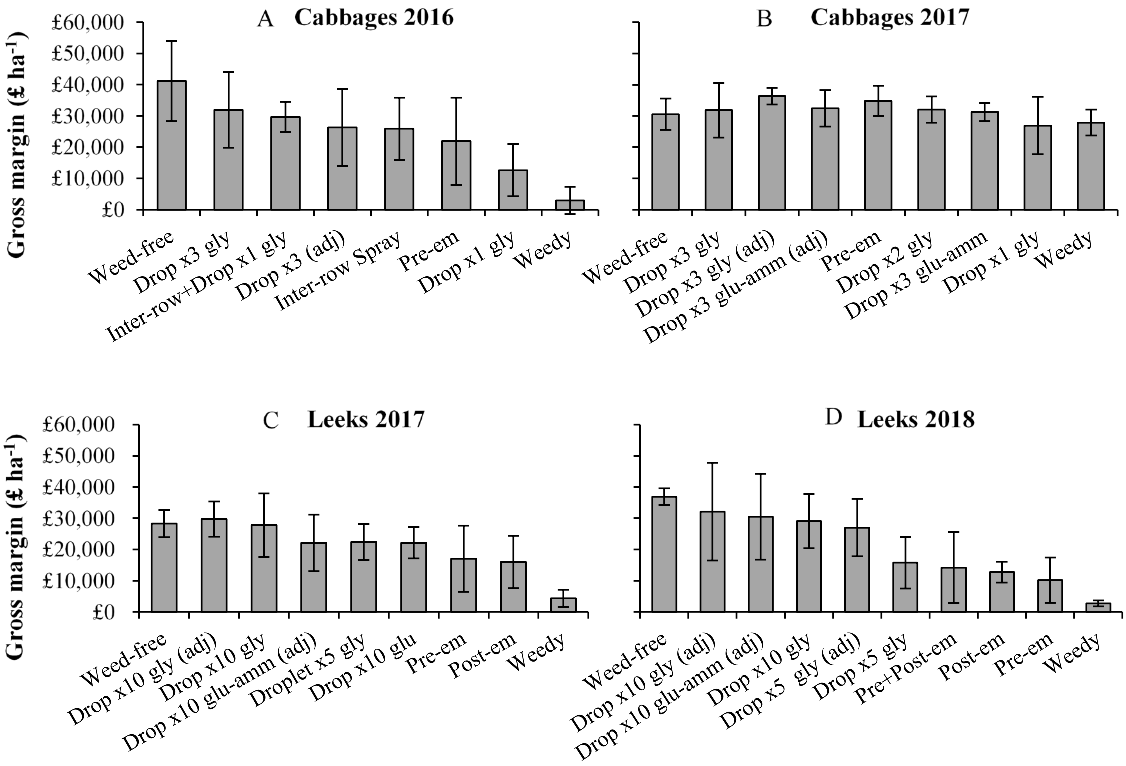


Figure 15 Gross margins (£ ha^{-1}) over costs of weed control for experimental weed control treatments applied to plots of cabbages in (A) 2016 and (B) 2017 and of leeks in (C) 2017 and (D) 2018. Droplets containing glyphosate (gly) or glufosinate-ammonium (glu-amm) were either applied plant-specifically (one droplet (Drop…) per plant) or leaf-specifically (one droplet per leaf (Drop (adj)), one to ten times (x1 - x10) during the growing season. Broadcast, pre- and post-emergence (Pre-em, Post-em) sprays were also tested. Values are means of three replicates (±SD). SEDs (df) are (A) £6,825 (21), (B) £3,430 (24), (C) £4,270 (16) and (D) £7,854 (18). (From Koukiasas et al. 2020).

the gross margins were used to predict the ***probability*** that a higher profit would occur using leaf-specific weed control compared to applying a conventional pre-emergence spray with pendimethalin. The results are presented showing two sigmoidal probability distributions. If the two weed control systems were similarly profitable, it would be expected that each distribution would cross the x-axis at the 50% level, such that there would be a 50% probability of making more profit and a 50% probability of making less profit. The slopes of the lines indicate the sensitivity and reflects the errors of measuring yields. According to the simulation, the grower of leek crops in these trials would have been 82 and 86% more likely to make a higher profit by controlling weeds leaf-specifically in 2017 and 2018, respectively (Figure 16). Or, one could also say that the simulation implies that 82-86% of leek growers would have been likely to produce higher profits. The rest of the curve is also useful. For example, there is a 50% probability of making an £18000/ha increase in profit and a 60% probability of £10000/ha more profit (Figure 16). The corollary also applies: up to 18% might make less profit.

For cabbages, the lack of significant differences in gross margin in 2017 (Figure 15 B) is also reflected in the Monte Carlo simulation with a much shallower curve and the break-even point being at a 38% probability for leaf-specific control with glyphosate and 60% for glufosinate ammonium (Figure 17). The take-home message for cabbage growers would be that even after all costs are included, their profitability is unlikely to be greatly affected but environmental benefits are considerable.

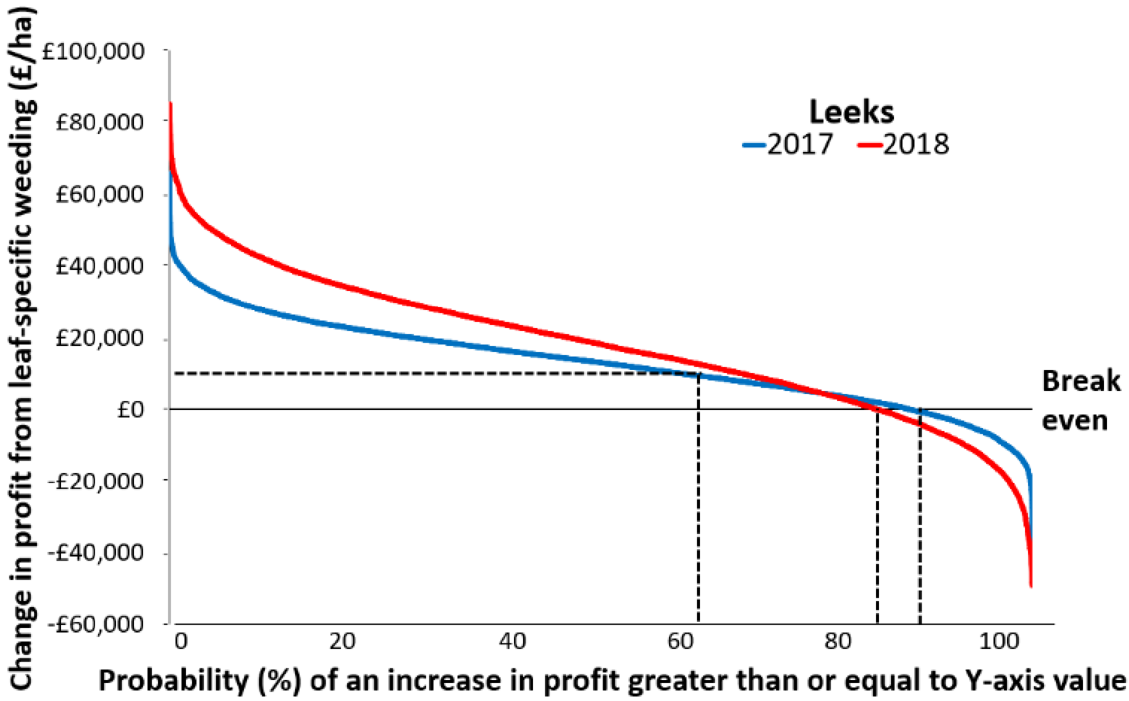


Figure 16 Predicted probabilities of an increase/decrease in profit from leaf-specific weed control compared to the profit from conventional pre-emergence spraying for leeks in 2017 and 2018. (Based on Monte Carlo simulation.)

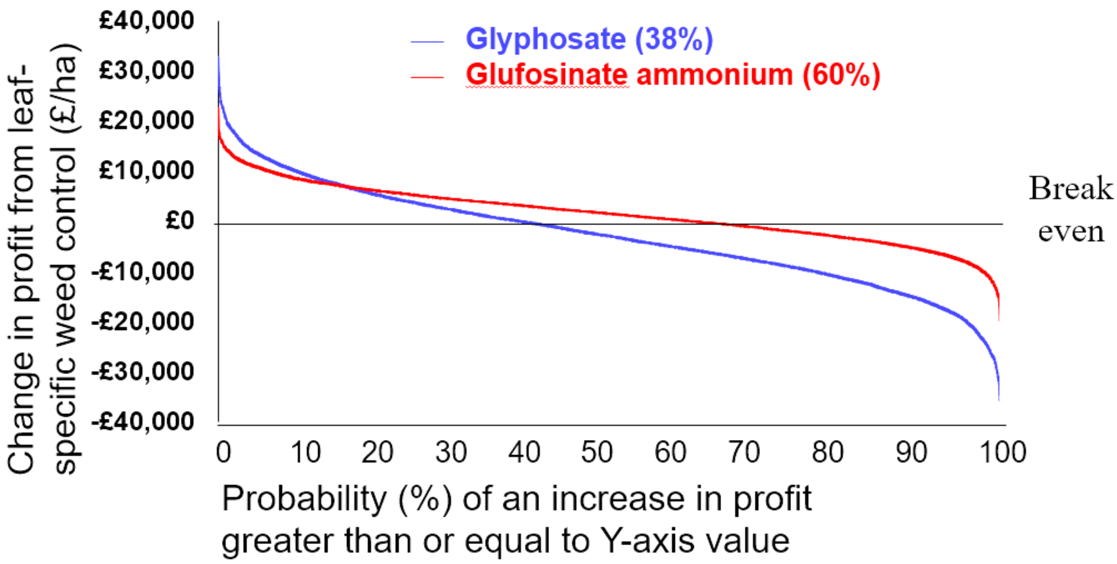


Figure 17 Predicted probabilities of an increase/decrease in profit from leaf-specific weed control with either glyphosate or glufosinate ammonium compared to the profit from conventional pre-emergence spraying for cabbages in 2017. The break-even probabilities are indicated in brackets. (Based on Monte Carlo simulation.)

Due to uncertainty regarding the platform cost, and the number of days per week when it could be operating, a sensitivity analysis was conducted to evaluate the effects of these variables on crop gross margins. With one exception, reductions in gross margins (Figure

15) when using glyphosate and glufosinate-ammonium droplets, associated with an increase in the cost of the platform from £25,000 to £100,000 per unit and a decrease in the operating days from seven to one day per week, were less than 0.7% and 4.8% for cabbages and leeks, respectively (Appendix Table 15). The exception was when the platform was operating one day per week, a larger decrease in the gross margins (2.3% and 18% for cabbages and leeks) was observed with increased cost of the platform (Appendix Table 15).

Preliminary economic analysis suggested that if the platform's cost is £25,000 and droplet treatments are carried out 5 days per week, the system would have been less profitable than weed-free control. On this basis, only when droplets of glyphosate were applied on ten occasions for the 2017 trial was the droplet treatment more profitable than the weed-free control. Gross margins of weekly applications of glyphosate droplets greatly exceeded those of conventional pre-emergence spraying, by £12,980 ha^{-1} and £22,255 ha^{-1} in 2017 and 2018, respectively (Appendix **Table 14**). Similarly, high values of additional profit were observed when applying droplets of glufosinate-ammonium compared with pre-emergence spray (up to £20,617 ha^{-1}). For cabbages just one droplet treatment (Drop x3 gly (adj)) was more profitable when compared with spray applications of pendimethalin in both years (2016: £4,521 ha^{-1} and 2017: £1,489 ha^{-1}) (Appendix **Table 14**). Droplet treatments appeared to be more profitable only for 2017 trial when compared to hand-weeding the cabbages. Based on the material cost of the glyphosate when using droplet applications, savings of up to £13 ha^{-1} and £19 ha^{-1} in herbicide costs were achieved when compared with spraying the pre-emergence pendimethalin for leeks and cabbages respectively (Appendix **Table 14**).

Discussion

Glasshouse dose-response trials

Results clearly showed that weeds can be controlled effectively using herbicide droplets which are applied leaf-specifically in glasshouse conditions. Droplet volumes (0.7 µl to 3 µl) were much larger than those produced by broadcast sprayers and concentrations of active ingredient(s) in the droplets varied from 2.5% to 20%. While uptake and translocation may, therefore, have been affected, the only visual evidence of a problem was when very high concentrations killed leaves at the point of droplet application, which would limit translocation.

Moreover, the log-logistic model, which is normally used to quantify dose-response relationships for conventionally-applied herbicide sprays, satisfactorily fitted the biomass data reported in most cases and control efficacy was generally excellent and as expected such that the calculated recommended rates achieved high efficacy. Encouragingly for glyphosate dose-response trials, the slope parameter (*b*) ranged from 1 - 3.5, which is similar to the range of 2 – 4 reported for conventionally-sprayed glyphosate (Streibig & Green, 2017). One exception was for glufosinate-ammonium droplet application to *P. annua* for which the lack of fit test was significant.

The need to evaluate alternatives to glyphosate is essential not only to reduce the risk of herbicide resistance, but also, the possibility of a loss of its approval as a plant protection product. Its use is permitted in the EU and the UK until 15 December 2022 and an application for renewal of this approval is in progress (EU, 2020 online). Thus, the efficacy of both glufosinate ammonium and 2,4-D as well as of co-applications of 2,4-D with either glyphosate or glufosinate ammonium confirmed that alternatives might be available for leaf-specific treatments should herbicide resistance arise or if the approval for glyphosate were removed. Interestingly, for *C. album*, the proprietary mixture of glyphosate with 2,4-D proved 3.3-times more potent than glyphosate alone. The search for other products should, however, be continued although detailed enquiries of two major agro-chemical companies, who fully understood this project, indicated that they were not aware of any suitable compounds from their chemical screening activities. We specifically asked about potential active ingredients where the production costs for broadcast spraying might be prohibitive, but given a 90+% reduction in herbicide required per hectare, a much more expensive product could still cost growers less per hectare if applied leaf-specifically.

With regards to the amount of active ingredient required to control individual seedlings using droplets of glyphosate, Mathiassen *et al*. (2016) reported that 3.7 µg of glyphosate per seedling reduced *C. album* seedling fresh biomass by 50%, which is of the same order as the results here for dry biomass. Utstumo *et al.* (2018) found that doses of 7.56 µg glyphosate per plant satisfactorily controlled *C. album* and *P. annua* in glasshouse conditions although dose-reponse curves were not assessed and the trial was not reproducible. Although direct comparisons are impossible due to differences in the sizes of seedlings, it is encouraging that these results are of the same order of magnitude as the ED_{50} estimates of 1.39 and 6.27 µg glyphosate per seedling observed here (**Table 10**: Trials 1(a) and 2(b).

In general therefore, weeds can be controlled leaf-specifically by droplet application to one leaf providing that non-selective, broad-spectrum, translocated herbicides are used. Moreover, for broad-leaved weeds, weed control can often be achieved with much lower doses than the label rates used for conventional spraying.

From the perspective of traceability, an important feature of leaf- and plant-specific weed control is that the herbicide weight applied to each individual plant can be recorded and geo-referenced whereas, when broadcast spraying, only the average application rate over large land areas is known. Moreover, applying single droplets to each weed has the potential to save both the amount and cost of herbicide used – and these savings compared to broadcast spraying are considered in the next section.

Field experiments

Corroborating the results in the glasshouse, the concept of applying micro-doses of herbicides using droplets applied plant- or leaf-specifically has also been demonstrated for fields growing cabbages and leeks. The primary hypotheses were accepted for both crops since the efficacy of weed control and the yield produced from the droplet treatments were not statistically significantly lower than the weed-free controls and were higher than current spraying methods.

For cabbages when micro-doses of glyphosate were applied either as a single or multiple droplets per weed on three occasions, over 90% weed control was achieved without any significant yield penalty. The hypothesis that multiple treatments would be needed was also accepted since a single treatment using glyphosate droplets gave poorer levels of weed control and yielded even lower than the weedy controls in both years. Theoretically, a single weed control treatment between three and five weeks after planting should have been enough to avoid yield loss, and, for example, Roberts *et al.* (1976) found that, for summer seeded cabbage in the UK, a single weeding three weeks after crop emergence was sufficient. Roberts *et al.* (1976) and Weaver (1984) both agreed that cabbages do not have a critical period of weed control. Melander *et al.* (2015) came to a similar conclusion in mechanical inter-row weeding trials.

Why then, may more than one droplet treatment be necessary? All the above studies utilize physical, mechanical or chemical weed control methods and total weed control was achieved when they are applied. However, a single leaf-specific application of herbicides as tested in this study is not expected to achieve 100% efficacy for two reasons. First, an important aspect of applying droplets to weed leaves is to avoid any herbicide reaching the crop either directly or by run-off after rain or as leaves move in the wind. Weeds growing in an area less than 1 cm from the edge of the crop were therefore deemed unsafe to treat due to a risk of collateral damage. Secondly, to avoid accidental direct applications to soil, very small seedlings (leaf area <1 cm^2) were also left untreated on the basis that they might not be targeted accurately enough by an automated system. Therefore, multiple treatments were required to control weeds that were omitted on the first visit. A third reason was to ensure effective weed control if weeds were poorly controlled or emerged after previous applications. So even though cabbages do not have a critical period for weed control, it is recommended that herbicide droplets should be applied on three occasions between 3 and 7 weeks after transplanting as demonstrated here.

Unlike cabbages, leeks have a clear critical weed-free period and in order to avoid yield losses of over 5% in leeks, the crop needs to remain weed-free during the critical weed-free period from 1 to 12 weeks after planting (Tursun *et al.* 2007). Not surprisingly therefore, multiple treatments using droplets of glyphosate and glufosinate-ammonium were found necessary to keep the crop largely weed-free. For both years, weekly applications of glyphosate droplets applied to every leaf starting from 3 and continuing until 12 weeks after planting, achieved at least 91% weed control efficacy without significantly lower yields. Also, a fortnightly application of the same treatment for 2018 controlled 100% of the weeds however, it did yield significantly lower than the hand-weeded, weed-free controls. A possible reason for that could be an infestation with leek miner which resulted in overall lower leek yields for the 2018 trial. Efficacy of weed control also appeared satisfactory by applying a single glyphosate droplet per weed (36 µg) on a weekly basis however yields for both years were significantly lower.

The amounts of herbicide a.i. applied in the optimal droplet treatments for cabbages (Drop x3 gly and Drop x3 gly (adj)) were 91% to 98% lower than for the conventional pre-emergence spray of pendimethalin. For leeks, applications of droplets containing glyphosate or glufosinate-ammonium demonstrated that herbicide inputs can be reduced by up to 82% and 74% respectively. So, the hypothesis that targeted droplet applications would significantly reduce herbicide inputs was accepted. From a regulatory perspective, the amounts of glyphosate applied in cabbages over the growing season (28.1 g ha^{-1} and 83.3 g ha^{1}) were from 85% to 98% lower than the minimum label recommendation for spraying the herbicide (540 g ha^{-1}). When a DoD system (Asterix robot) was used for intra-row weed control in fields with carrots applying 2.1 µl droplets of glyphosate , the equivalent of 191 g glyphosate ha^{-1} was applied, which is 73 to 91% lower than the minimum and the maximum label recommendations (540 g ha^{-1} to 2,160 g ha^{-1}) (Utstumo *et al.*, 2018). In this study, for glufosinate-ammonium the amount applied (40.2 g a.i. ha^{-1}) was also much lower than the recommended rates of 450 to 750 g ha^{-1}. Although more droplets were applied to the fields with leeks, the amounts of herbicides remained within the range of recommended doses, if not lower. Only in the case of the single droplet per weed of glufosinate-ammonium for the 2017 trial with leeks, were the amounts applied (2120.5 g ha^{-1}) above the maximum recommendation of 1500 g ha^{-1} (two applications of 750 g ha^{-1}).

Franco *et al.* (2017) predicted that the potential savings in herbicides using a micro-spraying system would be £11-22 ha^{-1}. This study supports this prediction: compared to spraying the full dose of a commercial herbicide, weed control by droplet application demonstrated savings in herbicide costs of up to £19 ha^{-1} for cabbages and £13 ha^{-1} for leeks.

Would these reductions in costs and amount of herbicides applied justify the investment in an automated platform? For leeks, the preliminary economic analysis presented here predicted a very high increase in crop profitability associated with droplet applications of both glyphosate (£22,255 ha^{-1}) and glufosinate-ammonium (£20,617 ha^{-1}) compared with spraying commercial herbicides (pre-emergence). These differences are due to the high value of leeks and the lower yields with pre-emergence herbicides. For cabbages however, only the adjusted glyphosate treatment appeared to be profitable for both years when compared with pre-emergence spraying.

The sensitivity analyses of the foregoing economic analysis was particularly encouraging. The gross margins were relatively insensitive to changes in platform costs to values much higher than anticipated. For example, Miller et al.'s (2011, 2013) spot spraying system costs approximately £45,000 and the applicator envisaged here is likely to be priced similarly. Concerns about the number of operational days were also addressed and provided the machine could operate for more than one day per week, profitability is not likely to be seriously compromised.

Despite these positive outcomes of the sensitivity analysis, the message may still be an insufficient incentive for growers to adopt what is a paradigm shift in weed technology. The obvious criticism is that the inferences are based on averages whereas an investment decision needs to take account of the risk that the benefit may not be realised. How likely is it that a grower would achieve a higher profit by adopting a leaf-specific weed control system? The Monte Carlo simulation approach has seldom been used to predict probabilities of profit as an aid to decision support. According to the simulation, leek growers would have been over 80% likely to increase their profits by adopting a leaf-specific weed control system with a 60% probability of their profits increasing by at least £10000 per hectare. For cabbages, where treatments showed few significant differences in gross margin, the take-home message was still positive: their profits were likely to be similar whatever weed control system they used but environmental benefits of controlling weeds leaf-specifically would be considerable with reductions in herbicide inputs in excess of 90%.

Optimised droplet treatments achieved satisfactory weed control without any yield penalty while reducing the amounts of herbicide applied per hectare by 70-90% in field grown leeks and cabbages. And, because of their high value, droplet applications could increase profits by over £11,000 ha^{-1} and £1,500 ha^{-1} per year for leeks and cabbages, respectively.

Figure 18 Custom-built camera system mounted on the boom of a small-plot sprayer, capturing images of weeds and red cabbage crop (left). Example of an image of a weedy plot with red cabbages as captured by the camera (right). The hoops were to support fleece.

Prototype platform development

Image capture

To facilitate software development for automated image capture and analysis required in for a leaf-specific prototype platform development Images in natural weed infestations in the 2016 and 2017 leek and cabbage experiments at Sonning Farm were captured automatically using a customised camera and custom-built computer system (supplied by Concurrent Solutions llc). The fixed focus camera was attached to a small-plot sprayer boom one metre above the crop canopy on a tractor-mounted sprayer (***Figure 18*** left). Image capture took place approximately weekly in leek and cabbage field trials at Reading. The camera was orientated to look straight down on to the centre of the field plots from above in order to capture images as would be carried out using an autonomous platform (***Figure 18*** right). Images were captured every 5 seconds and the tractor round speed was 8 km h^{-1}. For cabbages in 2016, images were captured weekly starting three weeks after and finishing seven weeks after transplanting.

Some semi-manual image analyses of images were carried out at Reading by Fern Price-Jones, an undergraduate student who was funded for six weeks during the summer of 2016 as part of the Undergraduate Research Opportunities Scheme (UROP) at Reading. The tagging software tool (developed by Paul de la Warr at Reading) enabled the operator place different “tags” on the digital images according to plant species. Leaves of cabbages, leeks were distinguished together with individual leaves of the weeds, fat hen, corn spurrey, chamomile, groundsel and other weeds (Figure 19). The weeds mentioned were the main ones growing in the plot.

Figure 19 Examples of tagged images of Savoy cabbage (left) and of fat hen (right). Individual leaves are tagged with red dots while plant centres are tagged with blue dots.

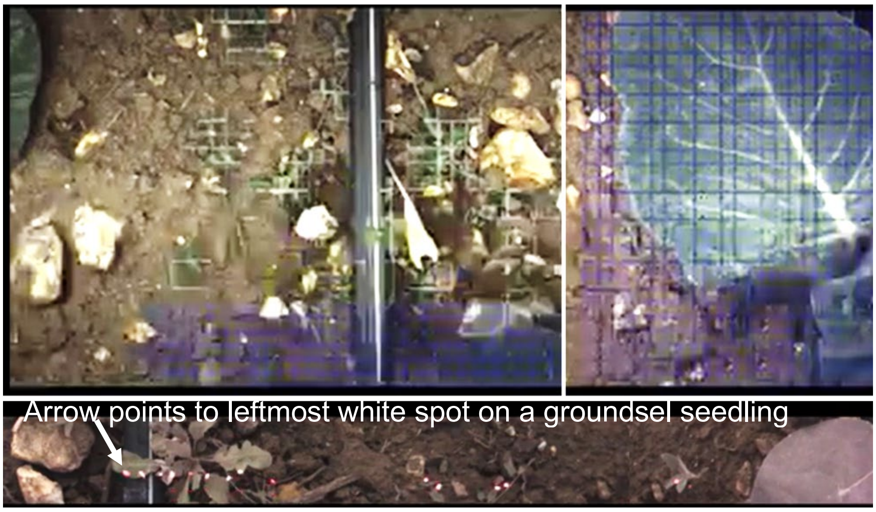


Figure 20 Images captured by prototype platform in cabbage plots at Sonning Farm, July 2019. The image was processed as 100 mm^2 squares. Blue shaded zones were protected to avoid crop damage (right). Squares with weeds are shown with white bounded squares (left and centre). Actual droplet targets are shown as white spots in the lower picture.

The tagged images were then available to Concurrent Solutions llc in the USA for developing the weed identification software for the prototype platform. In practice, although different weed species were tagged in the images, the identification of weed species is unnecessary for leaf-specific weed control using a broad-spectrum herbicide like glyphosate. The only classification needed is that between the crop and the weeds. All non-crop green leaves would be treated as weeds. Given this simplification and, because the crops were evenly spaced within each row and the row width was also constant, an important element in distinguishing the crop from the weeds was simply to identify the crop rows and crop plants within each row. Concurrent Solutions llc carried out further image capture in situ for the prototype platform demonstrated to growers in 2019 (Figure 20).

In 2019, a field day was organised by AHDB to demonstrate operation of a prototype autonomous platform (Figure 1) complete with an image analysis system linked to a single droplet applicator, in order to demonstrate the concept and technical feasibility and to assess interest from the industry.

Automated droplet applicator calibration and testing

Description

A prototype applicator system was designed and custom-built as an in-kind contribution for the eyeSpot project by Concurrent Solutions llc (**Figure 21**). Further development of a prototype robotic platform was also carried out by Concurrent Solutions llc. This unit was transported from Texas, USA, to Reading, installed and tested and then demonstrated at an AHDB Horticulture Open Day at Sonning Farm in July 2019. The remainder of this section describes calibration and targeting accuracy of the applicator system based on work carried out by in Benton, Kentucky by Nikolaos Koukiasas as part of his AHDB/Douglas Bomford Trust co-funded PhD project during summer 2018.

The system tested comprised a fluid application system and associated electronic controls. It was mounted on a miniature gantry, 1 m tall and 0.5 m wide. The applicator moved from right to left (**Figure 21**). The speed of this movement, the pneumatic pressure of the fluid system and the dispensing time for each droplet were all independently variable.

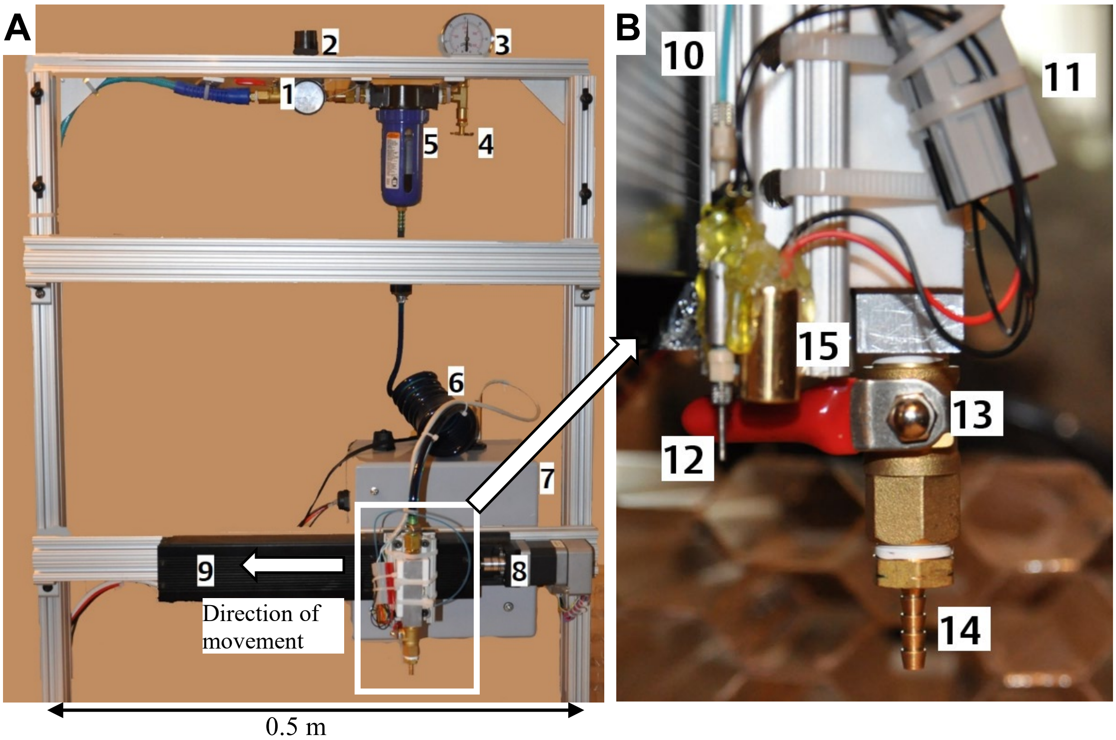


Figure 21. A) Front view of gantry system with B) enlarged view of applicator viewed in direction of motion. Numbered components are (1) air pressure shut off valve, (2) pressure regulator, (3) pressure gauge, (4) pressure release valve, (5) liquid reservoir, (6) flexible tubing, (7) controller box, (8) motor, (9) linear actuator, (10) ejector tubing, (11) manifold, (12) ejector nozzle, (13) drain valve, (14) liquid drain, (15) laser pointer. Pressure was delivered using a Husky 8G 150 PSI Hotdog portable Air Compressor.

Fluid pressure was varied by adjusting the air pressure from 0 to 60 psi with a pressure regulator (**Figure 21**). Droplets were dispensed by opening and closing a solenoid valve mounted before the nozzle with a minimum dispensing time of 1 ms enabling very fast cycling of the system as would be required for precise targeting of the leaves of small seedlings by a moving robotic platform carrying out weed control in vegetable fields. The solenoid valve and nozzle geometry were customised and purpose-built for the system and the specifications of these components are proprietary to Concurrent Solutions llc.

Calibration methodology

The time taken to dispense a 1 µl droplet was assessed at different pressures and valve opening times. The pressures tested were 69, 138, 207 and 276 kPa (10 to 40 psi) and the valve open times varied between 1 and 10 ms. Forty pressure/time combinations were tested with four repetitions. One thousand droplets of distilled water were dispensed into a 1.5 ml Eppendorf® microtube and the water was then weighed to the nearest 0.001 g. The average droplet weight was calculated by dividing this weight by 1000 and its volume estimated assuming a specific density of 1 g/mL.

Method of assessing targeting accuracy

Targeting accuracy of the moving and static applicator was tested with and without air movement. Targeting accuracy was measured by the displacement of the droplet on the paper target relative to the point of impact with a static applicator with no air movement. A fan was used to create air movement and wind speed was measured using an anemometer. A plastic honeycomb structure was used to reduce turbulence and maintain a constant wind direction. The target wind speed in all tests was 10 km h^{-1} and the aim was to achieve a similar windspeed and direction between the nozzle outlet and the target. Anemometer readings were therefore taken at the height of the applicator nozzle, halfway between the nozzle and the target and at the level of the target. Three wind directions were tested namely head (0°), cross (90°) and tail (180°) winds, angles being expressed relative to the direction of applicator travel. In every case, the effects on targeting accuracy of the distance of the nozzle from the target (up to 50 cm) and pressure (69 to 276 kPa), were assessed. The factorial experimental design became unbalanced because droplets fragmented such that displacement could not be measured when the nozzle was 50 cm from the target at 69 kPa pressure. A total of 99 different treatments were applied and the experiment was repeated four times. Ten droplets were applied to the target for each static treatment and five when the applicator was moving. When moving, there were six target locations per traverse, spaced at 3.9 cm intervals (Figure 22) over a total distance of 19.2 cm. The first and sixth targets of each traverse were ignored because the applicator was either accelerating or decelerating at these points (Figure 22). After allowing the droplets to dry, the application points were identified and the displacement from the control point with zero wind was measured for each wind direction.

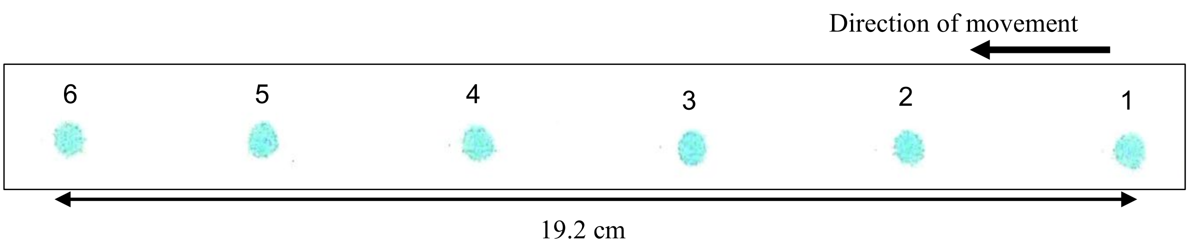


Figure 22 Droplet distribution when a single 1 µl droplet of coloured water was emitted from a moving applicator without any wind. The nozzle was 15 cm above the applicator, which was operating at 138 kPa. Note that no spatter occurred.

Results of calibration testing

A mean droplet weight of 1 µl was emitted when the applicator operated for 6, 4, 3 and 2.5 ms at pressures of 69, 138, 207 and 276 kPa, respectively (Figure 17). The unsurprising corollary is that the higher the pressure, the higher the flow rate through the nozzle and the greater the droplet volume for any given dispensing time (Figure 17). As might also be expected for a given pressure, there was a linear relationship between droplet volume and dispensing time meaning it would be easy to predict the dispensing times needed to apply a given droplet volume at a given pressure (Figure 17). The interaction of time and pressure on droplet volume was, however, significant ($P<0.001$) and this interaction is reflected in the curvilinear (logarithmic) relationship of the time and pressure required to emit a droplet of 1 µl (**Figure 24**).

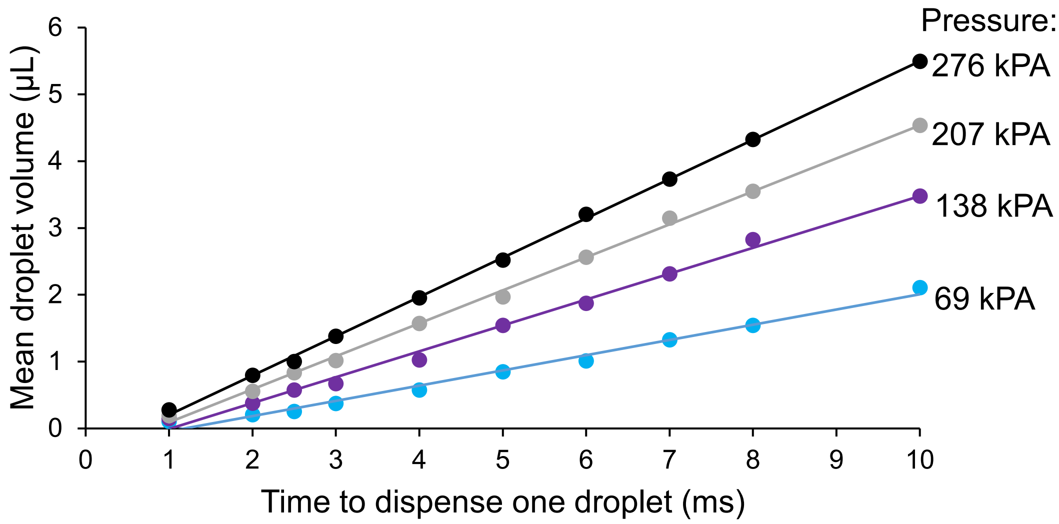


Figure 23 Droplet volume as a function of emission time at different pressures in the applicator. Volumes are means based on weights of 1000 droplets assuming a density of 1 g mL^{-1}. See Table 13 for parameter estimates of fitted lines.

*Table 13 Parameter estimates and standard errors (SE) of regressions (Figure 23**Error! Reference source not found.**) of droplet volume (µL) as a function of applicator emission time (ms) at different pressures.*

Pressure (kPa)	*Intercept*	*SE*	*Slope*	*SE*	*P-value*	R^2 correlation
69	*-0.270*	*0.024*	*0.228*	*0.004*	*<0.001*	*0.988*
138	-0.394	0.03	0.387	0.005	<0.001	0.994
207	-0.401	0.02	0.494	0.003	<0.001	0.998
276	-0.384	0.01	0.588	0.003	<0.001	0.999

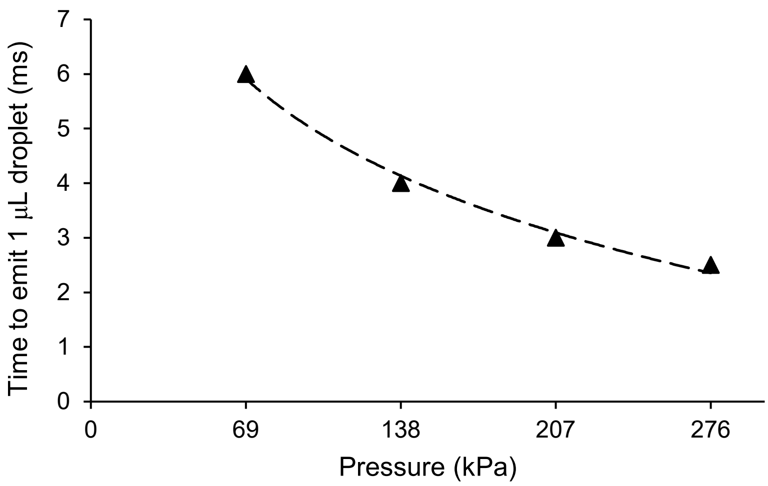


Figure 24. Effect of pressure (P) on the time (t) taken to dispense a 1 µL droplet. Fitted line (±SE of parameter estimates): t = -2.56 (±0.16) $\log_{10}(P)$ + 16.73 (±0.81) (R^2 = 0.99, p=0.002).

Targeting accuracy

The effects of distance of the applicator from the target, pressure, wind direction and motion of the applicator were investigated to optimise applicator performance in terms of minimum displacement from target and lack of droplet splitting.

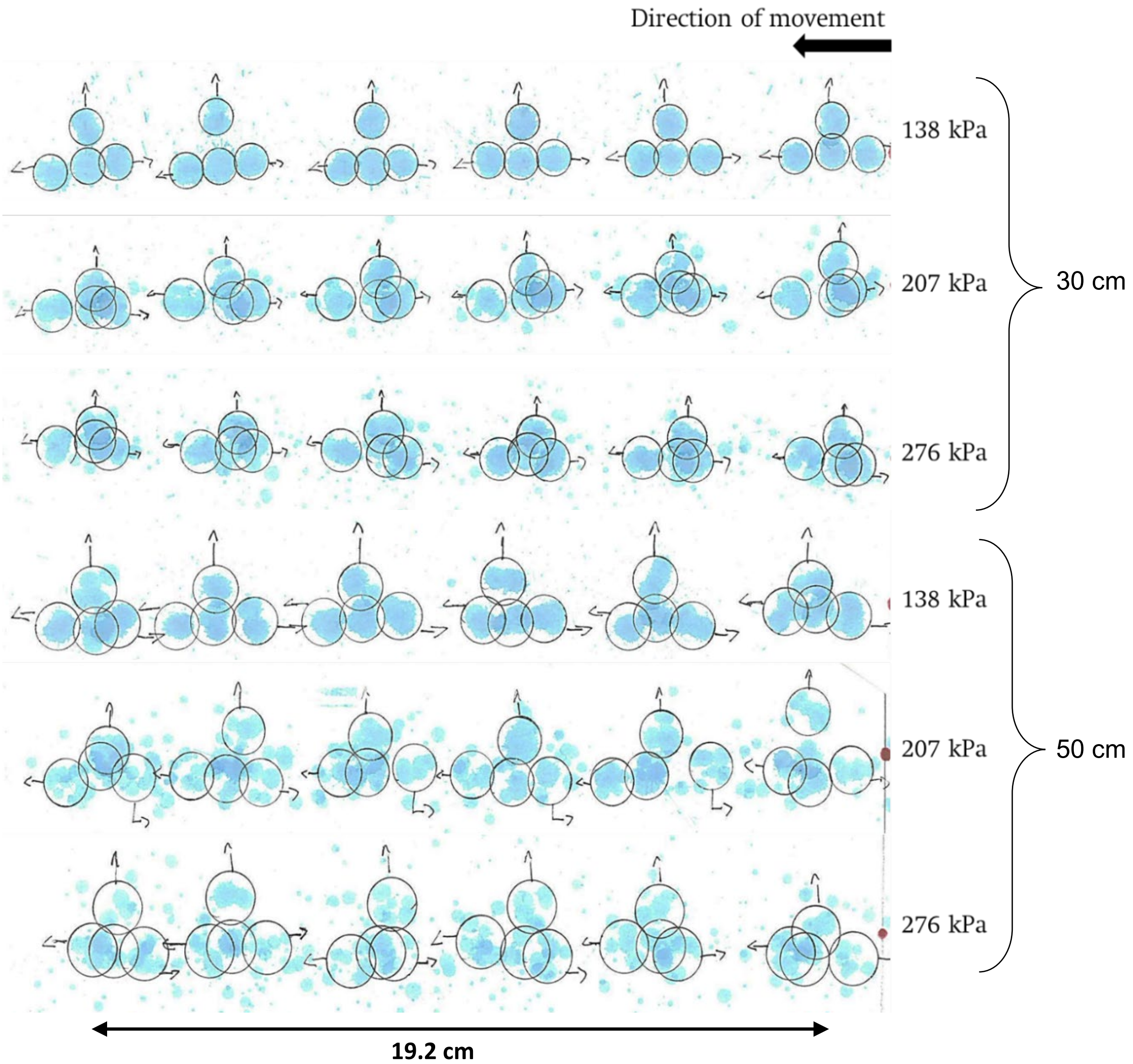


Figure 25 Results from moving applicator operating at 138, 207 and 276 kPa dispensing water droplets with blue dye from heights of 30 and 50 cm from the target and with side, head and tail wind directions. Circled areas indicate where five droplets were applied. Arrows on the circles indicate the wind direction (10 km h^{-1}) and also the direction of droplet displacement relative to the zero wind control (which has no arrow). Apparent spattering (microdroplets) is because gantry traversed five times over a short period of time and droplets were applied before the previous application had dried.

Visually, the greater the distance between the nozzle and the target and the lower the pressure, the greater the displacement due to wind (Figure 25). However, although the average displacement was therefore smaller at 276 kPa than with 138 kPa, the droplets emitted at both 207 and 276 kPa pressures showed a tendency to split (Figure 25

The smallest displacements were 1.8 to 4 mm and occurred observed when the nozzle was 15 cm above the target (**Figure 26**, **Figure 27**). Moreover, for this distance, the displacements did not differ significantly at pressures of 138, 207 and 276 kPa (**Figure 26**). Over all treatments, droplet displacement declined with increase in operating pressures. Moreover, the higher the distance from the target, the droplets tended to displace further. However, the largest displacement was observed when droplets were applied at 30 cm from the target and using 69 kPa pressure (**Figure 26** A).

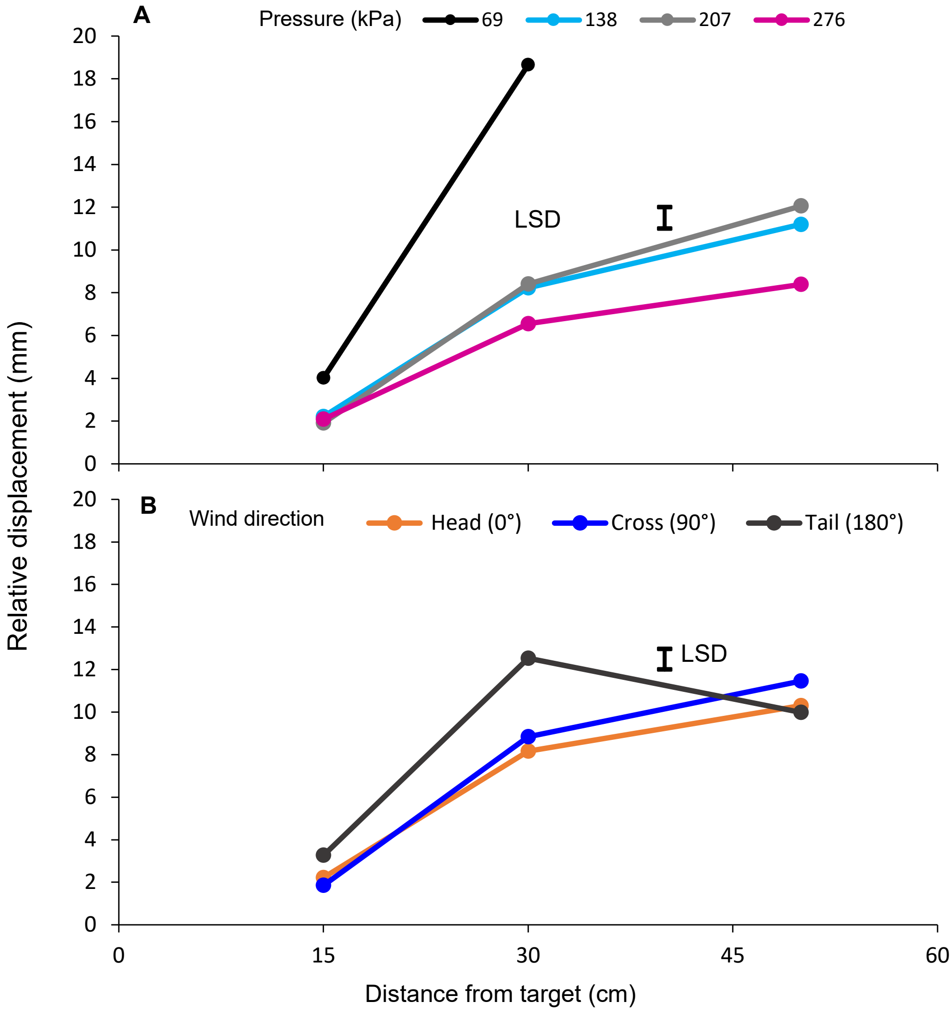


Figure 26. *Effect of distance from target (15, 30 and 50 cm) on the relative displacement of droplets relative to the zero-wind control, as a result of application of (A) four pressures (69, 138, 207 and 276 kPa) and (B) three wind directions (0°, 90° and 180°) regardless of the motion of the applicator. Maximum l.s.d. was 0.99 and 0.97 for A and B respectively (P=0.05).*

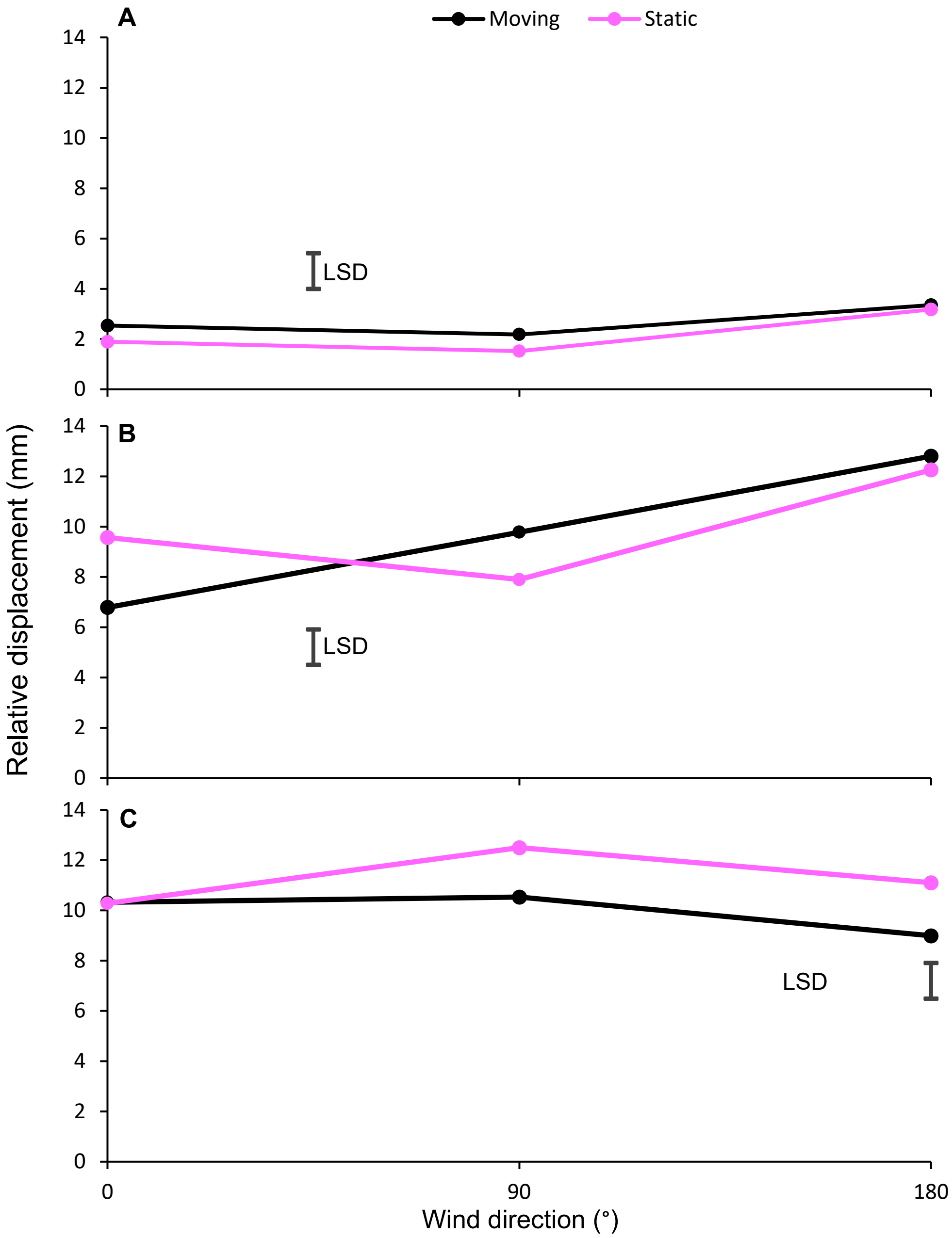


Figure 27. *Effect of 10 km h^{-1} wind speed when wind was applied from three different directions of 0° (head), 90° (cross) and 180° (tail) and distance from the target at (A) 15 cm, (B) 30 cm and (C) 50 cm to the displacement of the droplets when applicator was static and moving across the four pressures. Maximum l.s.d. is 1.41 (P=0.05).*

Wind direction was also important especially as the nozzle distance from target increased. A tail wind caused a 12.5 mm displacement of droplets emitted 30 cm from the target, significantly greater than that associated with head and cross winds at that distance. When the target was 50 cm from the nozzle, the displacement was not significantly different between tail and head winds (**Figure 26** B).

Overall, all variables tested – pressure, motion, distance and wind direction – affected targeting.

Given incidence of droplet splitting at 207 and 276 kPa, it was necessary to focus on an operating pressure of 138 kPa. At this pressure, the displacement caused by wind increased similarly with distance from the target when the applicator was moving or static (**Figure 28** B). Moreover, when the moving applicator was operating at 138 kPa, the displacements associated with wind direction did not differ significantly. For a static nozzle, the 7.3 mm displacement with a "tail" (180°) wind was only very slightly (1.26 mm) higher than with a head (0°) wind ($P<0.05$, Figure 28 A).

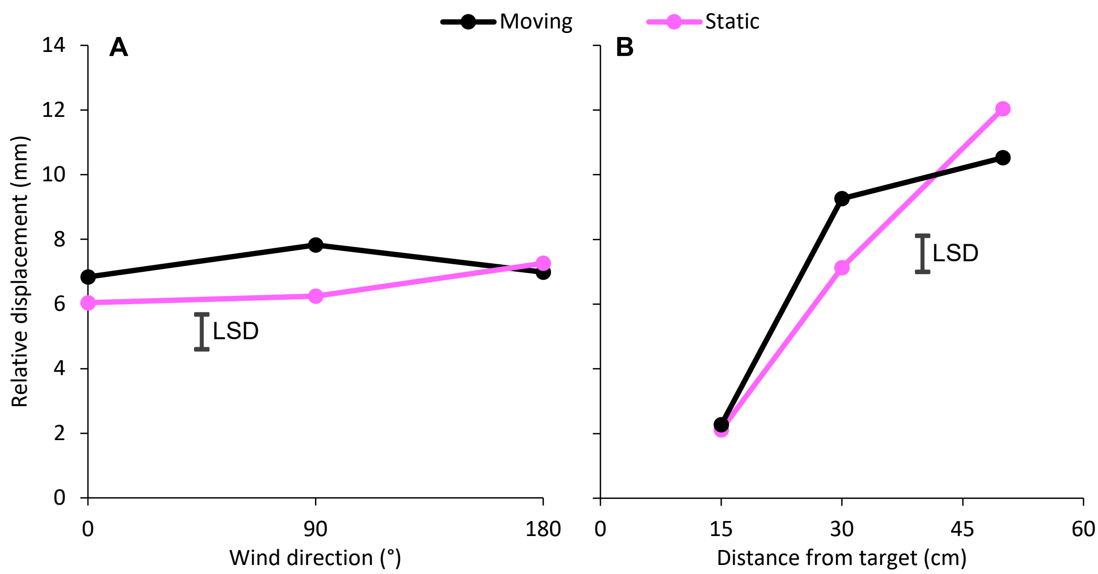


Figure 28. *Effect of (A) wind direction and (B) distance from the target on the displacement droplets from the no wind control, when applicator is operating at 138 kPa. Maximum l.s.d was 1.07 and 1.11 for (A) and (B), respectively.*

Discussion of applicator testing activities

A requirement for a practical working system in the field is that the dispensing time should not exceed 10 ms. The reason for this is based on the assumption in the economic analysis that the platform would treat 4 ha per eight-hour day. To achieve this with a platform with a swathe width of 1 m, the platform would need to travel at 5 km h^{-1}. At this speed, it would travel 14 mm every 10 ms. Given performance requirements that (i) leaves of 100 mm^2 (10x10 mm) should be successfully targeted and (ii) nozzles would not need to swivel in the direction of platform motion, it follows that the required dose would need to be dispensed in a period less than 10 ms in order for a correctly aimed, complete droplet to hit the leaf.

In the calibration tests, the time to emit a 1 μL droplet varied between 2.5 and 6 ms, the period decreasing with increase in pressure. This emission time would therefore be satisfactory for the required performance of the platform in the field. Moreover, given the use of a 2 m swathe in the prototype system demonstrated to growers in July 2019 as part of the project (Figure 1), there is clearly a considerable margin either to increase droplet size to 2 μL or to increase the hectarage covered per day. The attraction of a larger droplet size is to allow a lower concentration of herbicide to be used, reducing the risk of leaf scorch at the point of impact. By way of comparison with other experimental systems, a 10 ms spraying period 10 ms has been reported for micro-spraying (Lamm *et al*. 2002) and also for a droplet system (Søgaard and Lund 2007).

Higher pressures might be favoured not only because they reduce the emission time to achieve the required dose, but also because the displacement of droplets associated with wind and motion is reduced. The results clearly showed that the lowest operating pressure tested (69 kPa) could not be recommended for leaf-specific weed control due to the droplet displacement. However, the higher pressures of 207 and 276 kPa also proved unsatisfactory despite their potential for smaller displacements because the droplets split producing satellite droplets after emission leading to unpredictable and uncontrolled deviations from the target especially when the nozzle was 30 or more centimetres from the target. Given that a distinguishing requirement of the eyeSpot system is to hit very small seedlings, satellite droplets pose an unacceptable risk of missing the target and also of accidentally hitting the crop. Satellite droplets also occurred in Nieuwenhuizen *et al*.'s (2010) micro-sprayer and, when tested in the field to control volunteer potatoes, 1% of crop plants showed herbicide damage that may have been caused by these satellite droplets. Since it is necessary, therefore, for droplets to remain intact, an operating pressure of around 138 kPa should avoid the risk of satellite droplets in any future development of the prototype applicator system and platform (Figure 1) into a commercial machine.

To the risk of wind displacing the droplets, it is also clear that the closer the nozzle is to the target, the less the displacement. The question is: how low can you go? A 15 cm separation is conceivable in an experimental prototype and may be possible for peri-emergence weed control in a level field populated with small weed seedlings. However, transplanted crops such as the leeks tested in the field experiments, were up to 20 cm tall at planting, so that a nozzle-to-target separation of 30-40 cm may be required. In such a case, algorithms could be used to predict displacement and/or baffles could be installed as an engineering solution to minimise the impact of wind on displacement.

In conclusion, it is recommended that for use of the applicator in field conditions, the operating pressure should be set at around 138 kPa in order to avoid satellite droplets. Since the nozzle may often need to be more than 15 cm above the target, it is further recommended that options to mitigate droplet displacement due to wind should be fitted as standard equipment.

As a postscript, an appraisal of the potential for commercialisation was carried out by Concurrent Solutions llc and Knight Farm Machinery Ltd. after the Open Day in July 2019. It became clear that under present market conditions and following discussions with the industry, that the investment required would exceed the likely market for platforms in the UK for a field vegetable crop like leeks. Moreover, as has happened in the USA, well-financed corporate entities may invest large sums of money in these technologies making it difficult for small independent companies to be confident that they may achieve a return on the very substantial investment needed to bring a platform to market.

Conclusions

- It is useful to distinguish plant- and leaf-specific weed control. In the former, one droplet of herbicide is applied to each weed; in the latter, a droplet is applied to each leaf. In the eyeSpot project, plants or leaves with areas less than 100 mm^2 were generally left untreated to match the expected accuracy of targeting of an automated weeding system.
- The feasibility of leaf-specific weed control was clearly demonstrated and weeds up to the 4-leaf stage could be controlled satisfactorily by applying a droplet containing either 32 µg of glyphosate or 28 µg of glufosinate-ammonium to one leaf on each plant.
- In the field, the efficacy of weed control exceeded 90% with leaf-specific droplet treatments while simultaneously reducing the use of herbicide actives per unit land area by up to 82 and 94% in transplanted leeks and cabbages, respectively. The greater competitiveness of the cabbage crop reducing the number of droplet treatments and hence the amount of herbicide required to achieve a satisfactory efficacy of weed control.
- The economic analysis based on field experiments, predicted that weed control using **plant**-specific droplet applications to weeds in UK cabbage

crops and **leaf**-specific applications in UK leek crops would at least maintain and in many cases increase profitability of these crops for UK growers. These economic benefits include the full estimated costs of an automated system for droplet application. More specifically:

- In cabbages, three plant-specific droplet treatments with glyphosate droplets resulted in gross margins above the total costs of weed control of £32,000/ha for savoy cabbages in 2016, significantly higher than the £22,000/ha for conventionally-applied pre-emergence pendamethalin spray. Differences between weed control regimes were not significant in 2017 for white cabbage crops largely due to the greater competitiveness of the crop against weeds. Importantly, however, the plant-specific treatment did not significantly affect profitability even after allowing for additional machinery costs.
- In leeks, ten leaf-specific applications of glyphosate droplets at approximately weekly intervals in both 2017 and 2018 and a similar treatment using glufosinate-ammonium in 2018 achieved gross margins of £29-32,000/ha, much higher than the £10-17,000/ha for conventionally-applied pre-emergence pendamethalin spray.
- In order to support growers in deciding whether or not to adopt leaf- or plant-specific weed control, Monte Carlo simulation modelling predicted that there was over an 80% likelihood (probability) that a grower would make more profit by controlling weeds in leeks leaf-specifically compared to conventional spraying. Gross margins were predicted to be as much as £10,000/ha (per year) higher in 60% of cases.

- A prototype robotic platform for leaf- or plant-specific weed control in field vegetable crops in the UK, was specially designed, developed and built for the project by Concurrent Solutions llc. A working demonstration of this prototype aroused much interest from growers as well as the farming press especially as a result of the AHDB Twitter feed and an AHDB Press Release. The Open Day, held at Sonning Farm, Reading on 25 July 2019 presented the main results of the project, included plots showing the efficacy of leaf-specific weed control and showed the platform's ability to identify and target small individual weed seedlings safely without risking collateral damage to the crop.

- Overall, the precision farming concept of leaf-specific weed control as demonstrated and proven with the eyeSpot project's innovative, state-of-the-art system, would offer at least eight winning solutions for growers of field vegetables using. Were it to be commercialised, the eyeSpot platform could enable growers to:

  (i) maintain crop gross margins in cabbages or substantially increase them in leeks,

  (ii) achieve a high efficacy of weed control,

  (iii) potentially improve food quality by avoiding any herbicide application to the crop,

  (iv) reduce environmental impact by minimising non-target herbicide applications to the soil,

  (v) greatly reduce herbicide inputs per hectare,

  (vi) solve the problem of loss or lack of herbicide actives for field vegetables,

  (vii) address labour shortages for hand-weeding (non-organic) vegetables, and

  (viii) circumvent the need to breed herbicide-tolerant vegetable crops.

Knowledge and Technology Transfer

Videos (connected to Open Day 25 July 2019)

1. Collaborators, Concurrent Solutions llc, produced this video on the prototype robotic leaf-specific weed control platform for Open Day on 25 July 2019.
   261 views (13 August 2020)
   https://youtu.be/K-4_hdbNAes
2. AHDB video: “eyeSpot: a glimpse into the future of weed control”
   https://www.youtube.com/watch?v=pxIlvXc9REE
   1230 views (13/8/2020)
   There are links to this video from various farming press sources.
3. Twitter – 4 October 2019. “Meet eyeSpot” (@RobotAndAIWorld, @ahdb_hort 2061 views, 24 re-tweets, 55 likes) https://twitter.com/i/status/1180199835070291968
4.
   - AHDB video: “eyeSpot: a glimpse into the future of weed control”
     https://www.youtube.com/watch?v=pxIlvXc9REE

Press release September 2019 following Open Day on 25 July 2019

Hawk-eyed robot cuts chemical use by up to 95%

Research trials of a new automated weed-killing robot have reduced herbicide usage on crops by up-to 95 per cent.

A recent trial in Reading it was demonstrated how eyeSpot uses cameras to identify weeds in vegetable fields, eyeSpot then targets weeds individually and applies precise herbicide droplets with an ejector, which accurately fires treatment to individual leaves of each weed.

“This is precision agriculture in action, the robot has significantly reduced use of herbicides, while practically eliminating any harm to non-target organisms,” said AHDB Crop Protection Senior Scientist, Joe Martin.

Carried out at Reading University and part funded by AHDB, the research set out to protect the environment and help the industry manage with less access to crop protection products.

PhD researcher at the University of Reading, Nikolaos Koukiasas, who has been partly funded by AHDB and the Douglas Bomford Trust, said: “eyeSpot represents a paradigm shift to weed control by accurately targeting leaf-specific droplet applications. Preliminary results of manual droplet applications showed excellent weed control and a 95 per cent reduction of herbicide use in cabbages, and 74 per cent in leeks.”

With big data becoming instrumental in farming, eyeSpot's imagery also has the potential to be used for the observation of growth rates, enabling accurate scheduling of operations, early yield estimates and the detection of crop stress.

Alistair Murdoch, Professor of Weed Science at Reading, said: "Yields and profitability are likely to equal or exceed those achieved by conventional herbicide treatments without applying any chemical to the crop. The environment also benefits greatly by reducing the need for mechanical weed control, eliminating spray drift and reducing the possibility of chemicals entering the surrounding area."

Partners involved in the development of eyeSpot robot include Concurrent Solutions llc in the USA. The remainder of the project is supported by Knight Farm Machinery and is being partly funded at the University of Reading by the Douglas Bomford Trust and AHDB.

If successful in later stages of trials, the eyeSpot would be to serve the UK and potentially other worldwide markets. Opportunities for commercialization are being explored. <Ends>

eyeSpot media reports

1. 1 October 2014: AHDB Horticulture "CP 134 - "eyeSpot" – leaf specific herbicide applicator for weed control in field vegetables" https://horticulture.ahdb.org.uk/project/%E2%80%9Ceyespot%E2%80%9D-%E2%80%93-leaf-specific-herbicide-applicator-weed-control-field-vegetables [Sponsor's website notification]
2. 21 November 2016: AHDB Horticulture "The future of targeted weed control" https://horticulture.ahdb.org.uk/news-item/future-targeted-weed-control [Press report by sponsor]
3. 21 November 2016: Horticulture Week "Trials show promise of automated topical herbicide application" http://www.hortweek.com/trials-show-promise-automated-topical-herbicide-application/edibles/article/1416273
4. 21 November 2016: Fresh Produce Journal "New weed control tech 'could slash herbicide use'" http://www.fruitnet.com/fpj/article/170647/new-weed-control-tech-could-slash-herbicide-use
5. 21 November 2016: Farmers' Weekly "Precision spraying could reduce herbicide use by 95%" http://www.fwi.co.uk/arable/precision-spraying-reduce-herbicide-use-95.htm
6. 23 November 2016: Farmers' Guardian Insight. "Targeted weed control system under development".
7. 29 November 2016: Farming UK "New research project aims to reduce herbicide inputs in weed control by 95 per cent" [Published online 21 Nov 2016] https://www.farminguk.com/news/New-research-project-aims-to-reduce-herbicide-inputs-in-weed-control-by-95-per-cent_44865.html
8. 29 Nov 2016 Robotics cuts chemical use in vegetables | South East Farmer
9. 30 November 2016. "The future of targeted weed control". http://www.reading.ac.uk/apd/about/news/apd-news301116.aspx

10. 16 December 2016, News Report, “The future of targeted weed control” in Agroberichten Buitenland, Washington DC, USA. http://www.agroberichtenbuitenland.nl/verenigde-staten/future-targeted-weed-control/
11. 4 January 2017. *The Scottish Farmer*. Weed control by robot. http://www.thescottishfarmer.co.uk/news/15001410.Weed_control_by_robot/
12. 10 January 2017, News Report, “Robots: The Future of Weed Control” in AgWeb – the Farm Journal, Philadelphia, Pennsylvania, USA. http://www.agweb.com/article/robots-the-future-of-weed-control-naa-ben-potter/
13. 11 January 2017. “Robots: The Future of Weed Control – Tech Check News www.techchecknews.com/world/robots-the-future-of-weed-control/
14. 13 January 2017. “Can robots destroy weeds better than a farmer driving a sprayer?” In HortiDaily. Tholen, The Netherlands. http://www.hortidaily.com/article/31576/Can-robots-destroy-weeds-better-than-a-farmer-driving-a-sprayer
15. 13 Jan 2017 Robots: The Future of Weed Control | Weed Science Society of America wssa.net/2017/01/robots-the-future-of-weed-control/
16. 13 January 2017. “Researchers Developing Automated Spot Herbicide Ejector” In Growing Oregon. http://growingoregon.com/news/2017/01/researchers-developing-automated-spot-herbicide-ejector-2017-01-13/
17. Also in Growing Georgia http://growinggeorgia.com/news/2017/01/researchers-developing-automated-spot-herbicide-ejector-2017-01-13/
18. And in Growing Kansas http://growingkansas.com/news/2017/01/researchers-developing-automated-spot-herbicide-ejector-2017-01-13/
19. 18 January 2017. “Killing weeds is a job for robots” J-LYN Grains - Ontairo Farmer www.jlyngrains.com/ontairofarmer
20. 10 March 2017. “Reducing chemical inputs with robots” Rural News (New Zealand) https://www.ruralnewsgroup.co.nz/rural-news/rural-management/reducing-chemical-inputs-with-robots
21. 12 March 2017. “Can robots help reduce chemical inputs” FruitWorldMedia [quoting #15]
22. 13 August 2019. *SeedQuest.* United Kingdom - EyeSpot: a glimpse into the future of weed control. https://www.seedquest.com/news.php?type=news&id_article=109387
23. 14 August 2019. “eyeSpot – a glimpse into the future of weed control.” https://ahdb.org.uk/news/eyespot-a-glimpse-into-the-future-of-weed-control [AHDB news report headlining the Open Day “An exciting technological development in precision field vegetable production was demonstrated at Reading University’s Sonning Farm on 25th July.”
24. 14 August 2019. *Fruitnet.com / MACFRUIT Digital ON LINE 8-10 September.* (Ed Leahey) Weed bot cuts herbicide use by 95% http://www.fruitnet.com/fpj/article/179479/weed-bot-cuts-herbicide-use-by-95
25. 20 September 2019. “Precision herbicide applicator: Reading” https://ahdb.org.uk/precision-herbicide-applicator-reading [AHDB news release headlining the demonstration at Reading University’s Sonning Farm on 25th July.]
26. 20 September 2019. The eyeSpot project. Open Day Sonning Farm 25 July2019. [PowerPoint presentation slides]. https://projectblue.blob.core.windows.net/media/Default/Events/Horticulture/Event%20Docs/The%20eyeSpot%20project.%20Open%20Day%20Sonning%20Farm%2025%20July%202019.pdf

27. 3 October 2019. *FarmingUK*. Video: Weed-killing robot drastically reduces herbicide use. https://www.farminguk.com/news/video-weed-killing-robot-drastically-reduces-herbicide-use_54055.html
28. 3 October 2019. *Horticulture Week*. (Gavin McEwan) UK crop robot targets herbicide application while maintaining yield. https://www.hortweek.com/uk-crop-robot-targets-herbicide-application-maintaining-yield/fresh-produce/article/1661458
29. 3 October 2019. *Eurofruit*. (Fred Searle) Hawk-eyed robot 'cuts chemical use by 95 per cent'. http://www.fruitnet.com/eurofruit/article/179883/hawk-eyed-robot-cuts-chemical-use-by-95-per-cent
30. 4 October 2019. *AgriLand.* (Rachel Martin) UK weeding robot trials show chemical reduction of up to 95%. https://www.agriland.co.uk/farming-news/uk-weeding-robot-trials-show-chemical-reduction-of-up-to-95/
31. 4 October 2019. *Farmers' Guide*. Robot cuts chemical use by up to 95 per cent. https://www.farmersguide.co.uk/2019/10/robot-cuts-chemical-use-by-up-to-95-per-cent/
32. 13 October 2019. *Farmers' Weekly* (Johann Tasker) Tech Talk: Experts' verdict on the eyeSpot robot weeder. https://www.fwi.co.uk/machinery/technology/tech-talk-experts-verdict-on-the-eyespot-robot-weeder
33. 15 December 2019. *Doctoral Research Highlights 2019 – University of Reading.* Weed-killing robot that could transform weed control technology. Page 5 in: http://www.reading.ac.uk/web/files/graduateschool/Graduate_School_Highlights_2019_Web.pdf
34. 2020 (Undated). *AgriEXPO.* (Ben Potter). Robots: The Future of Weed Control. https://trends.agriexpo.online/project-61313.html
35. https://youtu.be/K-4_hdbNAes
    Twitter feed and videos from Open Day on 25 July 2019: #smarthort (then search for "eyespot" https://twitter.com/AHDB_Hort/status/1154383936358891525 (1.5 k views 13 August 2020)

Publications

1. Koukiasas N., Yu T. and Murdoch A. (2016). Dose response relationship of droplet applications for the leaf-specific weed control in vegetable crops. In: Proceedings of International Advances in Pesticide Application, Barcelona. *Aspects of Applied Biology,* **132,** 343-348.

2. Murdoch A., Koukiasas N., De La Warr P. and Price-Jones F. (2017). Precision targeting of herbicide droplets potentially reduces herbicide inputs by at least 90%. In: Proceedings Precision Systems in Agricultural and Horticultural Production, *Aspects of Applied Biology,* **135,** 39-44.

3. Koukiasas N, Martinez-Perez J, Pilgrim R, Sanford S, Murdoch A J. 2019. Implications of dose-response relationships of herbicide droplet applications for leaf-specific weed control in leeks. In: J V Stafford (ed.) Proceedings of The 12th European Conference On Precision Agriculture, 2019. *Precision Agriculture*, **19**, 209–215. Wageningen, The Netherlands: Wageningen Academic Publishers.

4. Murdoch, A.J., Koukiasas, N., Gadanakis, Y., Pilgrim, R.A., Sanford, S. (2019). Predicting the probability of higher profits by robotic weeding in leeks and cabbages in the UK. In: Abstracts of IOBC-WPRS Conference on Integrated Protection of Field Vegetables. Wellesbourne, U.K.: Association of Applied Biologists.

5. Koukiasas N., Gadanakis G., Pilgrim R., Sanford S. and Murdoch A. (2020). Predicting the profitability associated with robotic weeding in cabbages and leeks in the UK. In: Proceedings of International Advances in Pesticide Application, Brighton. *Aspects of Applied Biology*, **144**, 315-318.

Oral and poster presentations:

1. Alistair Murdoch Paul De La Warr, Nikolaos Koukiasas & Tzuyi Yu (Reading University), Carl Flint (Agrii, UK), Robert Pilgrim & Shane Sanford (Concurrent Solutions llc, USA), Brian Knight (Knight Farm Machinery Ltd, UK), Peter Lutman (UK), Paul Miller (NIAB/TAG, UK), Ben Magri & Tom Robinson (Syngenta, UK), Nick Walters (Patchwork Technology Ltd.) Automating site, plant and leaf-specific weed control in field crops: eyeWeed and eyeSpot. European Conference on Precision Agriculture, Tel Aviv, Israel, July 2015. Oral and abstract.
2. Koukiasas, N., Murdoch, A.J. Annual Weeds Review, Rothamsted, November 2015. Poster.
3. Koukiasas, N. Crops Production Symposium, Reading, November 2015. Oral.
4. Koukiasas, N., Yu, T., Murdoch, A.J. International Advances in Pesticide Application, Barcelona, January 2016, Oral & poster.
5. Koukiasas, N. SCI Young Researchers in Agri-Food, Reading, May 2016. Poster.
6. Koukiasas, N. SCI Young Researchers in Crop Sciences, July 2016. Oral.
7. Koukiasas, N., Murdoch, A.J. Annual Weeds Review, Rothamsted, November 2016. Oral & Poster.
8. Koukiasas, N. Crops Production Symposium, Reading, November 2016. Oral & Poster.
9. Koukiasas, N. AHDB Crops PhD Conference, Stratford-upon-Avon, November 2016. Oral & Poster.
10. Murdoch, A.J, Koukiasas, N. et al. FLRC Conference, New Zealand, February 2017. Oral.
11. Koukiasas, N. AgriFood Charities Partnership Student Forum, Hatfield, April 2017. Oral.
12. Koukiasas, N., Murdoch, A.J. eyeSpot meeting 16 August 2017. Link to presentation on Douglas Bomford Trust website: https://www.dbt.org.uk/sites/default/files/Eyespot%20Meeting%20August%20%2717.pdf
13. Murdoch, A.J, Koukiasas, N. et al. Precision Systems AAB Conference, Pershore College, October 2017. Oral.
14. Murdoch, A.J, Koukiasas, N. et al. British Onion Growers & British Leek Growers Meeting. Cranfield University, October 2017. Oral.
15. Murdoch, A.J, Koukiasas, N. et al. BCPC Annual Weeds Review. The eyeSpot Project. November 2017. Oral. Link to presentation on BCPC website: https://www.bcpc.org/events/reviews-events/bcpc-weeds-reviews/bcpc-weeds-review-2017
16. Koukiasas, N. Crops Production Symposium, Reading, November 2017. Oral & Poster.
17. Koukiasas, N. AHDB Crops PhD Conference, Stratford-upon-Avon, November 2017. Oral.
18. Koukiasas, N. Doctoral Research Conference, Reading, June 2018. Oral.
19. Murdoch, A., Koukiasas, N., Pilgrim, R., Sanford, S., De La Warr, P., Price-Jones, F. AAB Conference on Precision Systems in Agricultural and Horticultural Production. Pershore College, October 2017. Oral and paper.
20. Koukiasas, N., Murdoch, A.J., et al. Precision Spraying Event, Diss, November 2018. Oral
21. Koukiasas, N., Murdoch, A.J. et al. European Conference on Precision Agriculture. Montpellier, France, July 2019. Oral and paper.
22. AHDB Precision Weeding Open Day at reading, July 2019. Handout/PowerPoint presentation from Demonstration Day, 25 July 2019 is in Appendix.
23. Murdoch, A.J., Koukiasas, N., Gadanakis, Y., Pilgrim, R.A., Sanford, S. (2019). IOBC/AAB conference on Integrated Protection of Field Vegetables. Wellesbourne, U.K. Presented by N. Koukiasas. Oral and abstract.
24. Koukiasas N., Gadanakis G., Pilgrim R., Sanford S. and Murdoch A. (2020). International Advances in Pesticide Application, Brighton. Oral, Poster and Paper.

Educational impact and training

Dissertations and abstracts

Two MSc students successfully completed their Masters' dissertations and contributed significantly to the project (at no additional cost to the sponsors). Both have gone on to study for PhDs, Yu at Imperial College and Martinez at UEA. The project also sponsored Nikolaos Koukiasas as a PhD student. After completing his PhD, the project continued to employ him to help with writing up papers, complete supplementary data analyses and manage the demonstration for the Open Day. Since the completion of the project he has gone on to work for Syngenta at Jealotts Hill.

The titles and abstracts of these outputs are given below.

TzuYi Yu (2015) Understanding the dose response patterns and the related parameters of the organ-specific weed control approach in cabbage (*Brassica oleracea*) fields. Dissertation prepared in partial fulfillment of the requirement for MSc Agricultural and Development at the University of Reading.

Automated herbicide application on weeds using an organ-specific approach in a vegetable field was simulated in this research. Seedlings of the crop species (*Brassica oleracea* var. sabaud) and weed species (*Chenopodium album* and *Rumex crispus*) at Biologische Bundesanstalt, Bundessortenamt and Chemical (BBCH) growth stage 14–16, were treated with droplets of glyphosate with different doses and were harvested after 3 weeks. The associated experimental parameters and dose response curves were established and recorded. The ED_{90} values, assessed from shoot fresh weights of *B. oleracea*, *C. album* and *R. crispus* were 811, 32 and 353µg of glyphosate per plant, respectively. The ED_{90} values estimated from percentage reduction of dry biomass of *B. oleracea*, *C. album* and *R. crispus* were 345, 23, and 304µg of glyphosate per plant, respectively. Results indicate that applying the glyphosate containing herbicide by 32µg per plant at the post-emergence stage could effectively control the annul weed (*C. album*) without reducing the cabbage yields; however, the same dose would not be able to control the perennial weed (*R. crispus*). In addition, phytotoxicity assessments by European Weed Research Council (EWRC) scoring system were able to confirm and examine the dose response patterns. The results of this research are discussed with respect to further experiments for an automated system utilizing organ-specific weed control technology in field trials.

Javier Martínez Pérez (2018) Dose-response relationships of herbicide droplet application for leaf-specific weed control of *Chenopodium album*, *Poa annua* and *Senecio vulgaris*. Dissertation prepared in partial fulfilment of the requirements for the MSc. Agriculture and Development at the University of Reading.

Dose-response studies were carried out to test the efficacy of leaf-specific herbicide droplet application on three common weed species in vegetable crops. The herbicides used were: 2,4- D, glyphosate, glufosinate-ammonium and two combinations of them: 2,4-D + glyphosate and 2,4-D + glufosinate-ammonium. Herbicide droplets were applied to *Chenopodium album* (fat-hen), *Senecio vulgaris* (groundsel) and *Poa annua* (annual meadow grass) in glasshouse conditions. WinDias software was used to estimate the ground cover of the seedlings and the statistical package drc to estimate the herbicide required to decrease by 50% and 90% their dry biomass. *C. album* was controlled either with 2,4-D (1.1 µg/cm^2) or 2,4-D + glyphosate (2.7 µg/cm^2), applying 79% and 70% less active ingredient than their RD, respectively. *P. annua* was controlled with 2,4-D + glyphosate (17 µg/cm^2) or 2,4-D + glufosinate-ammonium (33.5 µg/cm^2), with an extra 44% or 60% more of active ingredient than the RD. *S. vulgaris* can be controlled with either glyphosate (0.1 µg/cm^2) or 2,4-D + glyphosate (0.6 µg/cm2) with 98% or 96% less active ingredient than their RD. It was proved that 2,4-D can control broadleaf weeds efficiently, but it is not recommended for *P. annua* weed control. 2,4-D was the best weed control option to control *C. album*. Although, 2,4-D + glyphosate was the best active ingredient combination to control all weeds tested. The combination of active ingredients can provide an alternative solution to manage weeds in a more sustainable way, saving up to 96% herbicide inputs when controlling weeds such as *S. vulgaris*. 2,4-D + glufosinate-ammonium had a synergistic effect controlling *P. annua* but antagonistic when applied on *C. album* or *S. vulgaris*. With this research experiment was demonstrated that leaf-specific weed control works under glasshouse conditions when used with common weeds within the UK between vegetable crops.

Nikolaos Koukiasas (2019) Leaf-specific weed control for vegetable crops in the UK. PhD thesis, School of Agriculture, Policy and Development, University of Reading.

Weed control in field vegetables in the UK is becoming increasingly challenging due to the loss of herbicide actives and demands by policy makers and consumers for lower pesticide use. Research at University of Reading in conjunction with Concurrent Solutions LLC in the USA, is developing a robotic weeder for field vegetables using image analysis to locate weed leaves and a novel Drop-on-Demand (DoD) applicator to apply droplets of herbicides to these leaves. No chemical is applied to the crop and none directly to the soil. Leaf-specific application of herbicide droplets is an alternative to selective chemistry or biotechnology while potentially reducing herbicide use. Although targeted micro-rates of herbicides have been studied, little is known about the exact rates needed to control weeds when microdoses are applied as one droplet to a single leaf or plant.

In glasshouse trials, individual weed seedlings were controlled by applying a single droplet of herbicide and dose-response relationships were quantified. As a general recommendation, weeds that are up to the 4-leaf stage can be controlled with a dose of 32 µg of glyphosate and 28 µg of glufosinate-ammonium when they are applied as a single droplet per seedling. In order to answer the question if the efficacy is reproducible in the field, manually applied droplets of glyphosate and glufosinate-ammonium were made to the naturally occurring weed population in transplanted cabbage and leek crop. Droplet applications made on three and ten occasions after transplanting the cabbages and leeks, respectively reduced residual weed biomass at harvest by over 90% compared to the weedy control. Also, droplet treatments gave a crop yield, which did not differ significantly from the weed-free control. At the same time, the total amount of herbicide active ingredient applied was up to 82% and 94% lower than currents spraying methods for the leeks and cabbages, respectively. Because of the high value of the crop and the higher yields associated with ultra-precise droplet application, it would appear to be economical to apply these droplets using a robotic weeder. The applicator which was developed by Concurrent Solutions LLC in the USA for Drop-on-Demand droplet applications was tested under indoor conditions. The effect of pressure, distance from the target, wind direction and motion of the applicator was tested on the targeting accuracy of the applicator. Recommendations for future field applications suggested that the applicator should operate at 138 kPa pressure and set at 15 cm height from weeds.

Higher Education curricular developments

Students at Reading, who are studying agriculture and agricultural business management, and MSc students studying a range of topics are all introduced to the eyeSpot technology as part of precision farming and crop agronomy teaching. Undergraduate and postgraduate students following an IPM module also gain practical experience and knowledge in a series of practicals in which they apply glyphosate leaf-specifically to weeds and analyse the results to produce dose-response curves as described in this report.

Reading University funds an undergraduate research opportunities (UROP) scheme giving undergraduate students the opportunity to obtain research experience on the project. We applied for this scheme and the student, Fern Price-Jones, assisted both in fieldwork and with image analysis, identifying the weeds and tagging them in the images. An example of her work is included in Figure 19.

Glossary

Leaf-specific weed control is where individual leaves are targeted to effect weed control.

Plant-specific weed control is where individual plants are targeted to effect weed control.

References

Baker S. & Knight J. (2017). What will happen to Plant Protection Product Regulation after Brexit? *Outlooks on Pest Management,* **28,** 93-94.

Bawden O., Kulk J., Russell R., McCool C., English A., Dayoub F., Lehnert C. & Perez T. (2017). Robot for weed species plant-specific management. *Journal of Field Robotics,* **34,** 1179-1199.

Blackmore S. (2013). In: Notes of a meeting, 16 July 2013, Digital Farming: The Next Agricultural Revolution, pp. 2-3. All-Party Parliamentary Group on Science & Technology in Agriculture, Palace of Westminster, London.

Blackmore S. (2014). Farming with robots 2020. In: Presentation delivered at Oxford Food Security Conference.

Bogue R. (2016). Robots poised to revolutionise agriculture. *Industrial Robot: An International Journal*, **43**, 450-456.

Brigham J. (2018). Wholesale fruit and vegetable prices. Available from: https://www.gov.uk/government/statistical-data-sets/wholesale-fruit-and-vegetable-prices-weekly-average [Accessed on 03/10/2019].

Christensen S., SøGaard H. T., Kudsk P., Nørremark M., Lund I., Nadimi E. S. & Jørgensen R. (2009). Site-specific weed control technologies. *Weed Research*, **49**, 233-241.

Ciba-Geigy (1975). Manual for field trials in plant protection. Ciba-Geigy, Basle, Switzerland.

EU (2020 online). Status of glyphosate in the EU. https://ec.europa.eu/food/plant/pesticides/glyphosate_en (accessed 22 July 2020).

Fennimore S. A., Cutulle M. (2019). Robotic weeders can improve weed control options for specialty crops. *Pest Management Science,* **75**, 1767–1774.

Fennimore S. A., Hanson B. D., Sosnoskie L. M., Samtani J. B., Datta A., Knezevic S. Z. & Siemens M. C. (2014). Field applications of automated weed control: Western hemisphere. Springer, Dordrecht, Netherlands, 151-169.

Franco C., Pedersen S. M., Papaharalampos H. & Ørum J. E. (2017). The value of precision for image-based decision support in weed management. *Precision Agriculture*, **18**, 366-382.

Garthwaite D., Barker I., Mace A., Parrish G., Ridley L., Macarthur R. (2017). Pesticide usage survey report 281-Outdoor vegetable crops in the United Kingdom. London, UK: DEFRA.

Hillocks R. (2012). Farming with fewer pesticides: EU pesticide review and resulting challenges for UK agriculture. *Crop Protection*, **31**, 85-93.

Jensen P. K. & Spliid N. H. (2003). Deposition of pesticides on the soil surface, Danish Environmental Protection Agency, Copenhagen, Denmark.

Koukiasas N. (2019). Leaf-specific weed control for vegetable crops in the UK. PhD thesis, School of Agriculture, Policy and Development, University of Reading.

Lamm R. D., Slaughter D. C. & Giles D. K. (2002). Precision weed control system for cotton. *Transactions of the Amarican Society of Agricultural Engineers*, **45**, 231.

Mathiassen S. K., Kudsk P. & Lund I. (2016). Adjuvants for single droplet application of glyphosate. In: Proceedings 2016 11th International Symposium on Adjuvants for Agrochemicals (eds ISAA Society) (20-24 June, Monterey, California, US). 131-137. Wageningen, Netherlands

Melander B. & Rasmussen G. (2001). Effects of cultural methods and physical weed control on intrarow weed numbers, manual weeding and marketable yield in direct-sown leek and bulb onion. *Weed Research*, **41**, 491-508.

Melander B., Lattanzi B. & Pannacci E. (2015). Intelligent versus non-intelligent mechanical intra-row weed control in transplanted onion and cabbage. *Crop Protection*, **72**, 1-8.

Midtiby H. S., Mathiassen S. K., Andersson K. J. & Jørgensen R. N. (2011). Performance evaluation of a crop/weed discriminating microsprayer. *Computers and Electronics in Agriculture*, **77**, 35-40.

Miller P.C.H., Lane A., Tillett N. & Hague T. (2010). Minimising the environmental impact of weed control in vegetables by weed detection and spot herbicide application. Final Report R270. AHDB/HDC, Stoneleigh.

Miller P.C.H., Lane A., Tillett N. & Hague T. (2013). Reducing herbicide use in row crops with targeted application methods treating detected weeds in small patches or spots. Final Report R435. AHDB, Stoneleigh.

Nieuwenhuizen A., Hofstee J. & Van Henten E. (2010). Performance evaluation of an automated detection and control system for volunteer potatoes in sugar beet fields. *Biosystems Engineering*, **107**, 46-53.

R Development Core Team. (2014). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. 2013. ISBN 3-900051-07-0.

Redman G. (2017). The John Nix Pocketbook for Farm Management 2018. Melton Mowbray, UK: Agro Business Consultants Ltd.

Ritz C. (2010). Toward a unified approach to dose–response modeling in ecotoxicology. *Environmental Toxicology and Chemistry*, **29**, 220-229.

Ritz C., Baty F., Streibig J. C. & Gerhard D. (2015). Dose-response analysis using R. *PloS one*, 10.

Ritz C. & Streibig J. C. (2012 online). Dose response curves and other nonlinear curves in Weed Science and Ecotoxicology with the add-on package drc in R. www.bioassay.dk.

Roberts H., Bond W. & Hewson R. (1976). Weed competition in drilled summer cabbage. *Annals of Applied Biology*, **84**, 91-95.

Ruckelshausen A., Biber P., Dorna M., Gremmes H., Klose R., Linz A., Rahe R., Resch R., Thiel M., Trautz D. & Weiss U. (2009). BoniRob–an autonomous field robot platform for individual plant phenotyping. Proceedings 7th European Conference on Precision Agriculture, pp. 841-849.

Scholz C., Kohlbrecher M., Ruckelshausen A., Kinski D. & Mentrup D. (2014). Camera-based selective weed control application module (“Precision Spraying App”) for the autonomous field robot platform BoniRob. Proceedings International Conference of Agricultural Engineering, Zurich. Paper C0598, 8pp.

Osnabrueck / Competence CScholz et al 2014

Slaughter D. C., Giles D. K., Fennimore S. A. & Smith R. F. (2008). Multispectral Machine Vision Identification of Lettuce and Weed Seedlings for Automated Weed Control. *Weed Technology,* **22,** 378-384.

Murdoch A.J., Smith L., Yates C.M & Rehman T. (2001). Predicting the probability of profitable weed control – a more reliable tool for making decisions. Proceedings of the Brighton Crop Protection Conference - Weeds, 1: 461-466.

Søgaard H. T. & Lund I. (2007). Application accuracy of a machine vision-controlled robotic micro-dosing system. *Biosystems Engineering,* **96,** 315-322.

Sørensen C. G., Nørremark M., Jørgensen R. N., Jensen K., Maagaard J., Jensen L. A. (2007). Hortibot: Feasibility study of a plant nursing robot performing weeding operations. *American Society of Agricultural and Biological Engineers* part IV: Paper No. 077019.

Streibig J. C. & Green J. M. (2017). Dose, Drift, and Non-Target Organisms. In: *Pesticide Dose: Effects on the Environment and Target and Non-Target Organisms,* (eds S.O. Duke & P. Kudsk), 25-45, American Chemical Society Publications, Washington, DC, US.

Tursun N., Bükün B., Karacan S. C., Ngouajio M. & Mennan H. (2007). Critical period for weed control in leek (*Allium porrum* L.). *HortScience,* **42,** 106-109.

Utstumo T., Urdal F., Brevik A., Dørum J., Netland J., Overskeid Ø., Berge T. W., Gravdahl J. T. (2018). Robotic in-row weed control in vegetables. *Computers and Electronics in Agriculture* **154**:36–45.

Weaver S. E. (1984). Critical period of weed competition in three vegetable crops in relation to management practices. *Weed Research,* **24,** 317-325.

Woznica Z., Idziak R., Waniorek W. & Sip M. (2015). An optimized glyphosate formulation. Proceedings 17th European Weed Research Society Symposium, Montpellier, France, p. 56.

Appendices

*Appendix **Table 14.** Gross margins and herbicide material costs of droplet treatments applied in the field experiments (2016-2018) and net profit or loss (in red) relative to pre-emergence spray, pre+post-emergence spray and hand-weeding (weed-free) treatments. Gross margins were calculated on the basis that the applicator would cost £25,000 and could operate five days per week. Pre + Post-emergence spray was not applied to cabbages. Assumptions underlying herbicide costs are presented in **Table 7**. Gross margins are presented in Figure 15.*

Treatments	Gross Margin (£ ha^{-1})		Material cost (£ ha^{-1})		Difference in gross margin (£ ha^{-1}) of droplet treatments compared to: Pre-emergence		Weed-free (hand-weeded)		Pre+Post-emergence
(A) Leeks	**2017**	**2018**	**2017**	**2018**	**2017**	**2018**	**2017**	**2018**	**2018**
Drop x5 gly	22,628	15,885	11	9.2	5,478	5,744	-5,759	-20,961	1,572
Drop x5 gly (adj)	N.A.	27,116	N.A.	7.7	N.A.	16,974	N.A.	-9,731	12,802
Drop x10 gly	28,273	29,308	15	13	10,998	19,166	-240	-7,539	14,869
Drop x10 gly (adj)	30,255	32,396	7	6.1	12,980	22,255	1,743	-4,451	17957
Drop x10 glu-amm	22,602	N.A.	25	N.A.	5,327	N.A.	-5,911	N.A.	N.A.
Drop x10 glu-amm (adj)	22,586	30,758	13	8.6	5,311	20,617	-5,926	-6,089	16,319
SED (d.f.)	4,270 (16)	7,854 (18)	3.6 (8)	1.1 (8)	3,394 (8)	9,628 (8)	3,394 (8)	9,628 (8)	9,628 (8)
(B) Cabbages	**2016**	**2017**	**2016**	**2017**	**2016**	**2017**	**2016**	**2017**	**Not applicable**
Drop x1 gly	12,680	26,975	0.9	0.3	-9,197	-7,987	-28,481	-3,599	
Drop x2 gly	N.A.	32,074	N.A.	0.7	N.A.	-2,748	N.A	1,550	
Drop x3 gly	32,065	31,795	1.4	0.9	10,188	-3,027	-9,097	1,271	
Drop x3 gly (adj)	26,398	36,311	2.1	0.6	4,521	1,489	-14,764	5,787	
Drop x3 glu-amm	N.A.	31,235	N.A.	1.5	N.A.	-3,586	N.A.	712	
Drop x3 glu-amm (adj)	N.A.	32,407	N.A.	0.8	N.A.	-2,415	N.A.	1,883	
SED (d.f.)	6,825 (21)	3,430 (24)	0.3 (6)	0.4 (15)	7,444 (6)	3,387 (15)	7,444 (6)	3,387 (15)	

N.A. Not Applicable

Appendix Table 15. Gross margins (£ ha^{-1}) of the glyphosate and glufosinate-ammonium droplet treatments used for the sensitivity analysis. Gross margins per hectare of conventional spraying treatments were £21,877 and £31,061 for the cabbages 2016 and 2017, respectively and £17,025 and £14,189 for the leeks 2017 and 2018, respectively. Glufosinate-ammonium was not applied in the 2016 trial with cabbages

			Gross margin, £ ha^{-1}	
			Treatments	
Crop	**Platform cost, £/year**	**Operating days/week**	**Glyphosate droplets**	**Glufosinate-ammonium droplets**
Cabbage 2016	5,000	1	31,815	N.A.
	10,000		31,565	
	15,000		31,315	
	20,000		31,065	
	5,000	3.5	31,994	
	10,000		31,922	
	15,000		31,851	
	20,000		31,779	
	5,000	5	32,015	
	10,000		31,965	
	15,000		31,915	
	20,000		31,865	
	5,000	7	32,029	
	10,000		31,994	
	15,000		31,958	
	20,000		31,922	
Cabbage 2017	5,000	1	36,061	32,156
	10,000		35,811	31,906
	15,000		35,561	31,656
	20,000		35,311	31,406
	5,000	3.5	36,240	32,335
	10,000		36,168	32,263
	15,000		36,097	32,192
	20,000		36,025	32,120
	5,000	5	36,261	32,356
	10,000		36,211	32,306
	15,000		36,161	32,256

Crop	Platform cost, £/year	Operating days/week	Gross margin, £ ha^{-1} Treatments: Glyphosate droplets	Glufosinate-ammonium droplets
	20,000		36,111	32,206
	5,000	7	36,275	32,370
	10,000		36,240	32,335
	15,000		36,204	32,299
	20,000		36,168	32,263
Leek 2017	5,000	1	29,005	21,336
	10,000		27,755	20,086
	15,000		26,505	18,836
	20,000		25,255	17,586
	5,000	3.5	29,898	22,229
	10,000		29,541	21,872
	15,000		29,184	21,515
	20,000		28,826	21,157
	5,000	5	30,005	22,336
	10,000		29,755	22,086
	15,000		29,505	21,836
	20,000		29,255	21,586
	5,000	7	30,076	22,407
	10,000		29,898	22,229
	15,000		29,719	22,050
	20,000		29,541	21,872
Leek 2018	5,000	1	31,396	29,758
	10,000		30,146	28,508
	15,000		28,896	27,258
	20,000		27,646	26,008
	5,000	3.5	32,289	30,651
	10,000		31,932	30,294
	15,000		31,575	29,937
	20,000		31,217	29,579
	5,000	5	32,396	30,758
	10,000		32,146	30,508

Crop	Platform cost, £/year	Operating days/week	Gross margin, £ ha^{-1} Treatments Glyphosate droplets	Glufosinate-ammonium droplets
	15,000		31,896	30,258
	20,000		31,646	30,008
	5,000	7	32,467	30,829
	10,000		32,289	30,651
	15,000		32,110	30,472
	20,000		31,932	30,294

N.A. Not Applied

Statistical analyses of field experiments

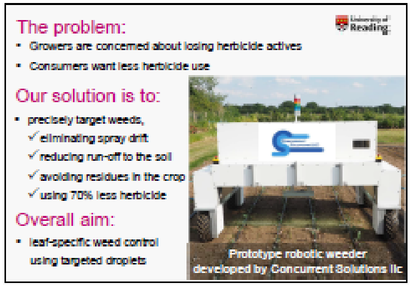
Appendix Table 16 One-way ANOVA analysis of trimmed and marketable yield, harvest index, reduction of weed biomass and rejection rate of unmarketable savoy cabbage heads for the 2016 and white cabbage heads for the 2017 trials.

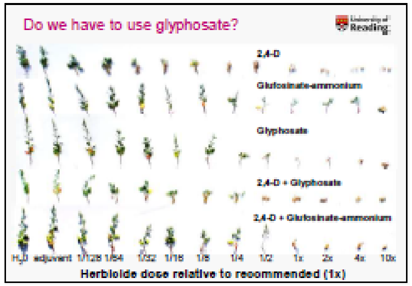
Year	Variable	Source of variation	d.f.	Sum of Squares	Mean Sum of Squares	*P*-value
2016	Relative trimmed yield (% of weed-free)	Blocks	3	5365	1788.3	
		Treatments	7	15565	2223.5	**0.006**
		Residual	21	11680	556.2	
	Relative marketable yield (% of weed free)	Blocks	3	9293.1	3097.7	
		Treatments	7	24016.1	3430.9	**0.01**
		Residual	21	19561	931.5	
	Economic value (£/ha)	Blocks	3	659,200,000	219,700,000	
		Treatments	7	4,090,000,000	584,300,000	**<0.001**
		Residual	21	1,956,000,000	93,160,000	
	Harvest index	Blocks	3	1000.9	333.6	
		Treatments	7	6428.9	918.4	**0.014**
		Residual	21	5671.4	270.1	
	Reduction of weeds' biomass relative to weedy control, (%)	Blocks	3	1097.5	365.8	
		Treatments	7	37386.8	5341.0	**<0.001**
		Residual	21	5616.0	267.4	
	Cabbage heads rejection rate, (%)	Blocks	3	483.4	161.1	
		Treatments	7	19917	2845.3	**<0.001**
		Residual	21	10336.9	492.2	
2017	Relative trimmed yield (% of weed-free)	Blocks	3	5767.2	1922.4	
		Treatments	9	3329.9	370.0	0.265
		Residual	27	7476.3	276.9	
	Relative marketable yield (% of weed free)	Blocks	3	14369.1	4789.7	
		Treatments	9	9164.3	1018.3	0.242
		Residual	27	19793.3	733.1	
	Economic value (£/ha)	Blocks	3	321,800,000	107,300,000	
		Treatments	9	336,500,000	37,390,000	0.13
		Residual	27	582,500,000	21,570,000	
	Harvest index	Blocks	3	1078.4	359.5	
		Treatments	9	2114.2	234.9	0.082
		Residual	27	3204.9	118.7	
2017	Reduction of weeds' biomass relative to weedy control, (%)	Blocks	3	2444.7	814.9	
		Treatments	9	35820.3	3980.0	**<0.001**
		Residual	27	13632.7	504.9	
	Cabbage heads rejection rate, (%)	Blocks	3	1909.7	636.6	
		Treatments	9	4590.3	510	0.09
		Residual	27	7048.6	261.1	

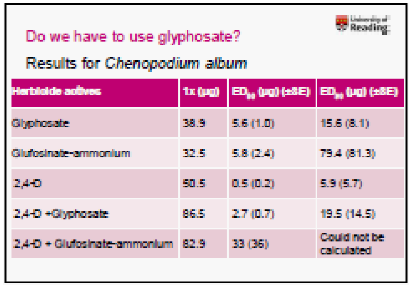
Appendix Table 17 Analyses of variance of trimmed marketable yield (% Weed-free), harvest index, economic value and reduction of weed biomass for the 2017 and 2018 trials with leeks.

Year	Variable	Source of variation	d.f.	Sum of Squares	Mean Sum of Squares	*P*-value
2017	**Relative trimmed marketable yield (% of weed-free)**	Blocks	2	4828.4	2414.2	
		Treatments	8	13673.2	1709.1	**<0.001**
		Residual	16	2900.3	181.3	
	Harvest index	Blocks	2	137.09	68.55	
		Treatments	8	134.38	16.80	0.860
		Residual	16	572.34	35.77	
	Economic value (£/ha)	Blocks	2	530,900,000	265,400,000	
		Treatments	8	1,918,000,000	239,700,000	**<0.001**
		Residual	16	437,400,000	27,340,000	
	Reduction of weeds' biomass relative to weedy control, (%)	Blocks	2	178.8	89.4	
		Treatments	8	37602.7	4700.3	**<0.001**
		Residual	16	1813.9	113.4	
2018	**Relative trimmed marketable yield (% of weed-free)**	Blocks	2	752.6	376.3	
		Treatments	9	23126.1	2569.6	**<0.001**
		Residual	18	2966.3	164.8	
	Harvest index	Blocks	2	22.77	11.38	
		Treatments	9	184.44	20.49	0.73
		Residual	18	554.33	30.80	
	Economic value (£/ha)	Blocks	2	63,760,000	31,880,000	
		Treatments	9	4,264,000,000	473,800,000	**0.002**
		Residual	18	1,666,000,000	92,530,000	
	Reduction of weeds' biomass relative to weedy control, (%)	Blocks	2	1500.6	750.3	
		Treatments	9	30507.9	3389.8	**<0.001**
		Residual	18	6452.8	358.5	

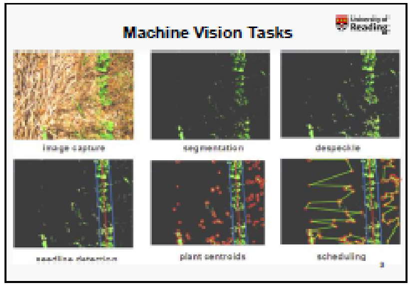
Handout/PowerPoint presentation from Demonstration Day, 25 July 2019

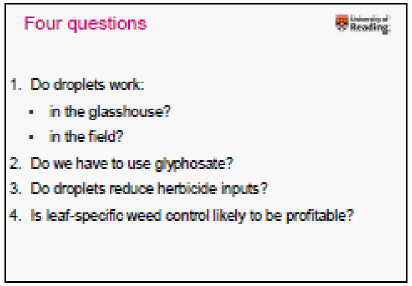


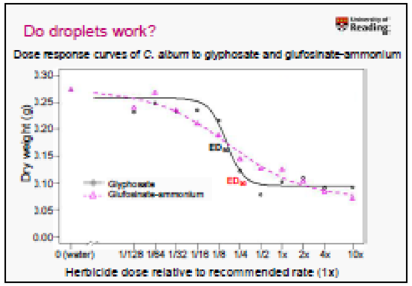


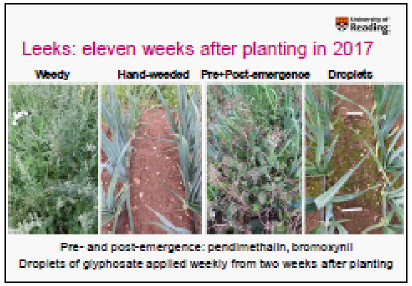


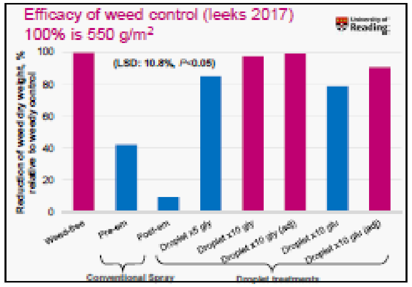
Herbicide actives	1x (µg)	ED50 (µg) (±SE)	ED90 (µg) (±SE)
Glyphosate	38.9	5.6 (1.0)	15.6 (8.1)
Glufosinate-ammonium	32.5	5.8 (2.4)	79.4 (81.3)
2,4-D	50.5	0.5 (0.2)	5.9 (5.7)
2,4-D +Glyphosate	86.5	2.7 (0.7)	19.5 (14.5)
2,4-D + Glufosinate-ammonium	82.9	33 (36)	Could not be calculated

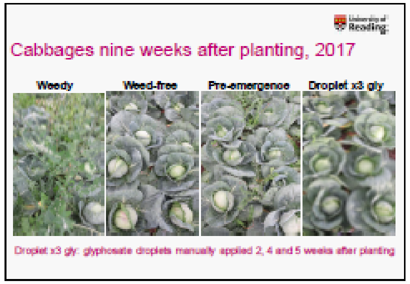


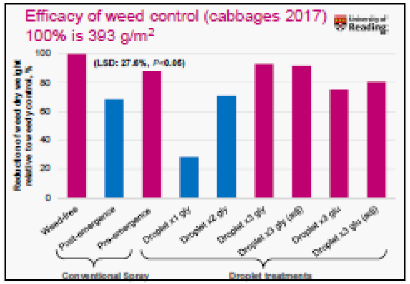


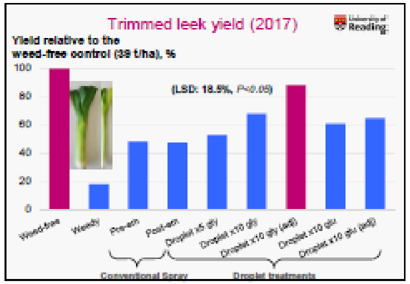


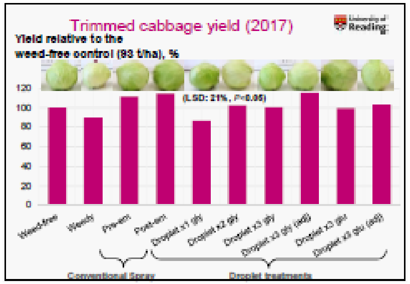


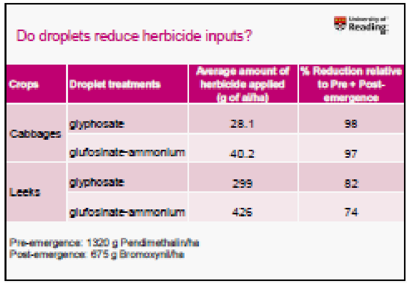


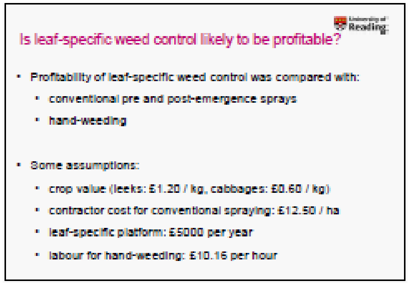

University of Reading
Cabbages nine weeks after planting, 2017
Weedy
Weed-free
Pre-emergence
Droplet x3 gly
Droplet x3 gly: glyphosate droplets manually applied 2, 4 and 5 weeks after planting

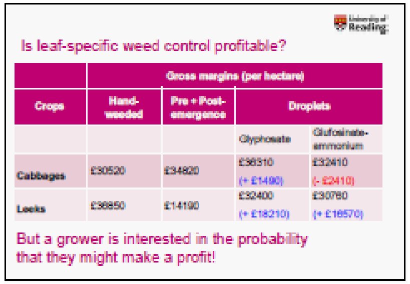

University of Reading
Efficacy of weed control (cabbages 2017)
100% is 393 g/m²
Reduction of weed dry weight relative to weedy control, %
(LSD: 27.8%, P<0.05)
100
80
60
40
20
0
Weed-free
Post-emergence
Pre-emergence
Droplet x1 gly
Droplet x2 gly
Droplet x3 gly
Droplet x3 gly (adj)
Droplet x3 glu
Droplet x3 glu (adj)
Conventional Spray
Droplet treatments

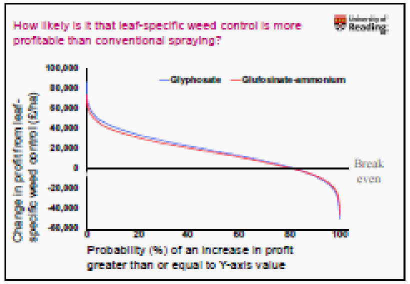

University of Reading
Trimmed leek yield (2017)
Yield relative to the weed-free control (38 t/ha), %
(LSD: 18.5%, P<0.05)
100
80
60
40
20
0
Weed-free
Weedy
Pre-em
Post-em
Droplet x5 gly
Droplet x10 gly
Droplet x10 gly (adj)
Droplet x10 glu
Droplet x10 glu (adj)
Conventional Spray
Droplet treatments

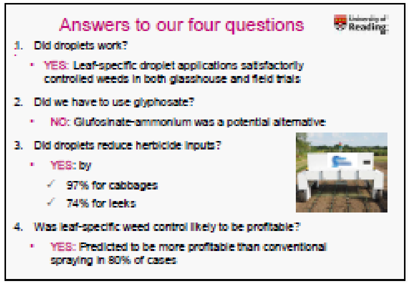

University of Reading
Trimmed cabbage yield (2017)
Yield relative to the weed-free control (93 t/ha), %
(LSD: 21%, P<0.05)
120
100
80
60
40
20
0
Weed-free
Weedy
Pre-em
Post-em
Droplet x1 gly
Droplet x2 gly
Droplet x3 gly
Droplet x3 gly (adj)
Droplet x3 glu
Droplet x3 glu (adj)
Conventional Spray
Droplet treatments

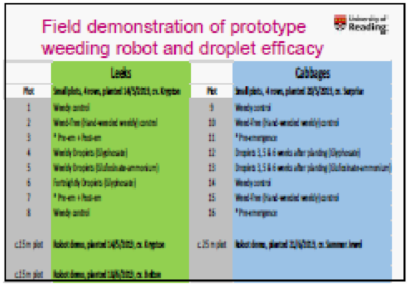

University of Reading
Do droplets reduce herbicide inputs?
Crops | Droplet treatments | Average amount of herbicide applied (g of ai/ha) | % Reduction relative to Pre + Post-emergence
Cabbages | glyphosate | 28.1 | 98
glufosinate-ammonium | 40.2 | 97
Leeks | glyphosate | 299 | 82
glufosinate-ammonium | 426 | 74
Pre-emergence: 1320 g Pendimethalin/ha
Post-emergence: 675 g Bromoxynil/ha


University of Reading
Is leaf-specific weed control likely to be profitable?
Profitability of leaf-specific weed control was compared with:
conventional pre and post-emergence sprays
hand-weeding
Some assumptions:
crop value (leeks: £1.20 / kg, cabbages: £0.60 / kg)
contractor cost for conventional spraying: £12.50 / ha
leaf-specific platform: £5000 per year
labour for hand-weeding: £10.16 per hour


University of Reading
Is leaf-specific weed control profitable?
Gross margins (per hectare)
Crops | Hand-weeded | Pre + Post-emergence | Droplets
Glyphosate | Glufosinate-ammonium
Cabbages | £30520 | £34820 | £36310 (+ £1490) | £32410 (- £2410)
Leeks | £36850 | £14190 | £32400 (+ £18210) | £30760 (+ £16570)
But a grower is interested in the probability that they might make a profit!


University of Reading
How likely is it that leaf-specific weed control is more profitable than conventional spraying?
Glyphosate
Glufosinate-ammonium
Change in profit from leaf-specific weed control (£/ha)
100,000
80,000
60,000
40,000
20,000
0
-20,000
-40,000
-60,000
Break even
0
20
40
60
80
100
Probability (%) of an increase in profit greater than or equal to Y-axis value


University of Reading
Answers to our four questions
1. Did droplets work?
YES: Leaf-specific droplet applications satisfactorily controlled weeds in both glasshouse and field trials
2. Did we have to use glyphosate?
NO: Glufosinate-ammonium was a potential alternative
3. Did droplets reduce herbicide inputs?
YES: by
97% for cabbages
74% for leeks
4. Was leaf-specific weed control likely to be profitable?
YES: Predicted to be more profitable than conventional spraying in 80% of cases


University of Reading
Field demonstration of prototype weeding robot and droplet efficacy
Leeks
Cabbages


University of Reading
Thank you for listening
Any questions?